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SOME PROBLEMS IN FUNGUS PHYLOGENY

ERNST A. BESSEY [1]

(WITH 5 FIGURES)

Owing to the nature of fungi their fossil remains are mostly limited to hyphae in fossilized wood and to the remains of pycnidia or perithecia in leaves or stems, with a few records of woody sporophores of polypores. These do not reveal much that is of help in phylogenetic studies, except to show us that higher fungi existed as parasites and saprophytes millions of years ago. Failing palaeontological records recourse must then be had to the other resources of the phylogeneticist: comparative morphology and ontogeny, and serum diagnostic studies.

Comparison of the morphology of now existing species is useful on the theory that evolution may result in divergences so that the more nearly related species will be those with the greatest similarities and, conversely, those that are less similar will represent greater evolutionary modifications and hence more distant relationship. This is probably true in the main, but there are many opportunities for error. It must be borne in mind that sometimes a single gene difference may produce a very great effect. Some dwarf plants, *e.g.* the Cupid sweet pea, appear to have arisen from large plants by this method and yet the difference is so striking that we would be inclined to consider the organisms as quite distantly related. So we may, on the basis of comparative morphology, erect a scheme of ascent or descent in which because of some

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such marked difference we may place two organisms far apart when perhaps they are really very closely related. The duplication of chromosomes, polyploidy, may also mislead us.

Another source of error is the frequently arising impossibility within a group of related organisms of deciding which characters are the more primitive and which the more advanced. In the Erysiphaceae the genera *Podosphaera* and *Sphaerotheca* produce but a single ascus in each perithecium, but in the very similar *Microsphaera* and *Erysiphe* respectively the asci are several to many, as is true of the other genera of the family. Upon examination of the closely related Meliolaceae and the less closely related Aspergillales we find that these are practically all polyascous so that we may safely conclude that the monascous condition in the Erysiphaceae is derived. Yet this does not grant us the right to conclude that this is necessarily true for the Ascomyceteae as a whole, even though, for other reasons, the speaker believes that the Saccharomycetales, which are monascous, are not primitively so but through reduction from groups with numerous asci. It is apparent therefore that the student of fungus phylogeny must know thoroughly not only the special group he is studying but all other groups that are possibly more or less closely related.

We cannot deny the possibility of convergent evolution. As an example take *Olpidiopsis* and *Pseudolpidiopsis* from the group formerly called the Chytridiales. Both arise from naked zoöspores which penetrate the host cells and enlarge there, sooner or later forming a cell wall. Two such adjacent cells may fuse, the contents of the one entering the other. Eventually zoöspores are produced which escape through an exit tube. Yet in *Olpidiopsis* the zoöspores are anteriorly biflagellate and the cell wall gives the cellulose reaction with chloriodide of zinc while in *Pseudolpidiopsis* the zoöspores are posteriorly uniflagellate and chloriodide of zinc does not call forth the cellulose reaction. We might conclude that these are very closely related organisms which have undergone relatively minor mutations as to flagellar number or that they represent converging branches of widely different groups of not closely related organisms. Another example, which clearly does not indicate close relationship, is *Tremellodon*, of the Order Tremellales, whose fruiting body bears its hymenium on teeth after the manner of

some species of *Hydnum*, one of the Eubasidiae. Here the differences in basidium structure and comparison with related forms make it certain that the similarity of the sporophores of the two organisms is pure convergence.

Carl Mez (**20**) has attempted to measure the degree of relationship by serum diagnosis methods. These are based on the theory that the genes are nucleoprotein units and that the more of the genes that are identical, *i.e.* the more closely related the organisms are, the greater will be the reaction. Thus the greater the precipitation produced in an animal serum sensitized by a given species of plant when nucleoprotein solutions from another plant are added, the greater the degree of relationship. The studies of Mez and his students have shown great correlation between the conclusions drawn from this method of study and the relationships assumed as a result of morphological studies. Yet in some instances the two methods have led to very divergent conclusions. How far may we trust either? Where both methods agree shall we say that one confirms the other? In that case which shall we consider the more trustworthy, the nucleoprotein reaction or the conclusion drawn from the actually visible results of the interactions of these proteins (genes)? Can we with fairness accept the confirmation of our phylogenetic scheme by serum diagnostic methods when they agree and reject them when they do not agree with our scheme as we have worked it out on the basis of comparative morphology, aided perhaps by our unrecognized preconceived ideas?

All of the foregoing reveals some of the mazes and pitfalls that lie before one when he attempts by one method or another or by a combination of methods to judge of relationships and of the probable course of evolution in so great a group of exceedingly varied organisms as the fungi.

In the following pages are discussed a few only of the many problems of fungus phylogeny. An attempt will be made to approach each problem from its various viewpoints without endeavoring to claim that the final solution has been reached.

The first problem is that of Slime Molds and their probable or possible allies. The organisms to be discussed have in common the fact that the encysted stage is the spore, the whole vegetative life cycle consisting of the naked amoeboid or plasmodial stages.

The true Slime Molds (Myxomycetes or Myxogastres of various authors, not in all cases with the same limits) and the Plasmodiophoraceae produce spores which possess cell walls of more or less disputed composition. Upon germination they set free usually one, sometimes more, naked, anteriorly flagellate cells more or less endowed with the ability to change their form in an amoeboid manner and to ingest solid food. The flagella are single or two in number. In the Plasmodiophoraceae they are always two and unequal in length. In the Slime Molds, proper, more frequently the flagellum is single, and anterior, but Gilbert (**12**) and later Sinoto and Yuasa (**27**, **34**) have shown that occasionally anteriorly biflagellate, but uninucleate cells are produced. These are not similar to the biflagellate zoöspores studied by Cotner in *Blastocladia,* which are binucleate and represent two cells that have failed to separate into the normal uninucleate, uniflagellate cells. In the biflagellate swarm cells of the Slime Molds the flagella may be equal or unequal, each flagellum arising from a minute granule just within the surface of the plasma membrane. In the uniflagellate cells which make up the majority of the swarm cells these two granules are present but from only one of them does a flagellum arise. From these circumstances it seems logical to assume that the ancestors of both these groups possessed anteriorly biflagellate swarm cells and that the Slime Molds have progressed further than the Plasmodiophoraceae toward the complete loss of one flagellum.

The Acrasiales and Labyrinthulales resemble the two foregoing groups in being naked except as to their spores. The latter, however, give rise to naked amoeboid cells without any flagellum in the Acrasiales, and in one genus of the Labyrinthulales, another genus of this latter group, *Labyrinthomyxa,* possessing one anterior flagellum. These naked cells are myxamoebae, like these into which the flagellate swarm cells of the two preceding groups become transformed. In the Acrasiales these naked myxamoebae draw together into easily separating masses or pseudoplasmodia, which, according to Skupienski (**28**), become true plasmodia in some cases. In the Labyrinthulales these myxamoebae join but become pushed apart by slender, thread like extensions forming a so-called net plasmodium with large lacunae. These cells divide by fission and draw apart with a connecting process. Eventually all

may round up and encyst, forming spores. Whether or not a sexual stage occurs has not been determined.

Apparently the ancestral forms were aquatic, naked, more or less amoeboid organisms with two anterior flagella and, like most naked Protozoa, with the power of encysting to survive unfavorable environments. Probably they easily united into plasmodial masses. The Acrasiales and the true Slime Molds have emerged to become aerial organisms, at least as to their fruiting structures, thus permitting aerial distribution of their spores. Connected with this is the production of the characteristic, often stalked sporangia with structures for support and for setting free the spores (stipe, columella, capillitium, peridium, etc.). These structures are lacking in the internally parasitic Plasmodiophorales and Labyrinthulales. Quite similar to the foregoing groups is the genus *Pseudospora* of Cienkowski. A zoöspore infects a host cell (usually of an alga) and develops into a plasmodium containing numerous nuclei. Eventually this breaks up into numerous naked, uninucleate, anteriorly uniflagellate more or less amoeboid, swarm cells which infect other host cells and thus complete the life cycle. Under certain conditions the whole plasmodium encysts but not the individual swarm cells. With the same life history is *Barbetia* of Dangeard (sometimes included in *Pseudospora*) in which the swarm cells are anteriorly biflagellate. These two genera are usually considered to be either pseudopodial Flagellata or flagellate *Rhizopoda.*

From their life history and structure it seemed logical to the great mycologist Anton de Bary (**3**), sixty years ago, to conclude that these groups of naked-bodied organisms, the Slime Molds and Plasmodiophorales, are not at all closely related to plants but that they are true animals. It must be noted that protozoölogists like Calkins (**7**), Kudo (**14**) and Minchin (**21**) include them in the group of Protozoa among the Sarcodina. Minchin classifies them as Phylum Protozoa, Class Sarcodina, Order Mycetozoa, dividing this order into Sub-order Euplasmodida (including the true Slime Molds) and Sub-order Sorophora (the Acrasiales), with *Labyrinthula* and *Plasmodiophora* as a closely related appendix but not definitely assigned to either sub-order. Kudo also places the Slime

Molds, and the other groups, along with *Pseudospora,* close together in the Sarcodina.

Some protozoölogists derive the Flagellata from Rhizopoda, regarding the flagella as especially modified pseudopodia. Others believe that the Flagellata are the more primitive, and that those

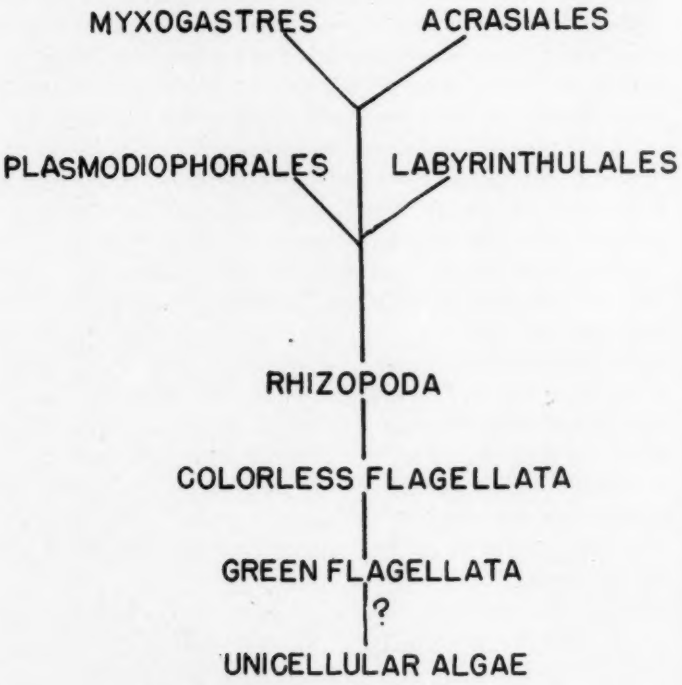

FIG. 1. Suggested phylogeny of Mycetozoa.

of this group that are amoeboid are more advanced on their way to the Rhizopoda with flagella and thence to those Rhizopoda lacking flagella. Carl Mez (**20**) has suggested that the chlorophyll-bearing Flagellata are derived from algae whose naked swarm-cell stage has become the predominant vegetative stage, the encysted stage being limited to the survival of unfavorable conditions. In the accompanying diagram (FIG. 1) this possible origin of the Flagellata and of the Mycetozoa is shown.

Turning away from the Animal Kingdom let us consider some organisms accepted by botanists as fungi, even though G. W. Martin (**19**) does not consider the fungi to be, properly, plants, but a third kingdom parallel to animals and plants and arising together with these in a common ancestral group of very simple organisms. Concerning the phylogeny of the organisms until recently included in the single order Chytridiales we find many conflicting ideas. Like the Mycetozoa the organisms of this group also possess a stage consisting of naked flagellate cells which may even show the ability to change their shape in an amoeboid manner. It is doubtful, however, whether they actually ingest particles of food as do the naked cells of the Slime Molds. The Chytridial swarm cells eventually encyst, externally to or within the substratum, and enlarge, becoming multinucleate and forming a sporangium directly or dividing into several sporangia. From these arise the swarm cells. Various types of sexuality are known, chiefly the union of two swarm cells or of two adjacent organisms.

In this group the zoöspores escape by the softening of the tip of an exit tube or papilla or by the formation of an operculum. This difference is being made use of in generic and family distinction by the more recent students of the group. The structure of the swarm cells, particularly the number, structure and location of the flagella is coming more and more to the fore in the systems of classification of these organisms. In the majority of the genera there is a single posterior flagellum of the whip-lash type, with no lateral tinsels. These form the Order Chytridiales in the narrower sense. Two or three genera, *e.g. Rhizidiomyces,* possess only one flagellum, and that in the anterior position. Couch (**8**) has shown this to be of the tinsel type. Usually also included in this order is the family Olpidiopsidaceae (formerly called the Woroninaceae, a name that may have to be abandoned if the genus *Woronina* is transferred to the Plasmodiophoraceae, as has been suggested). In this family the swarm cells have two flagella, at the anterior end of the cell or somewhat lateral. They are equal or one is somewhat longer, in that case always the anterior one if they are laterally attached. This flagellum is, according to Couch, of the tinsel type, the other being of the whip-lash type.

Correlated with the differences between one posterior and two anterior flagella are differences in the cell wall composition. In the Olpidiopsidaceae the cell wall responds immediately with the typical

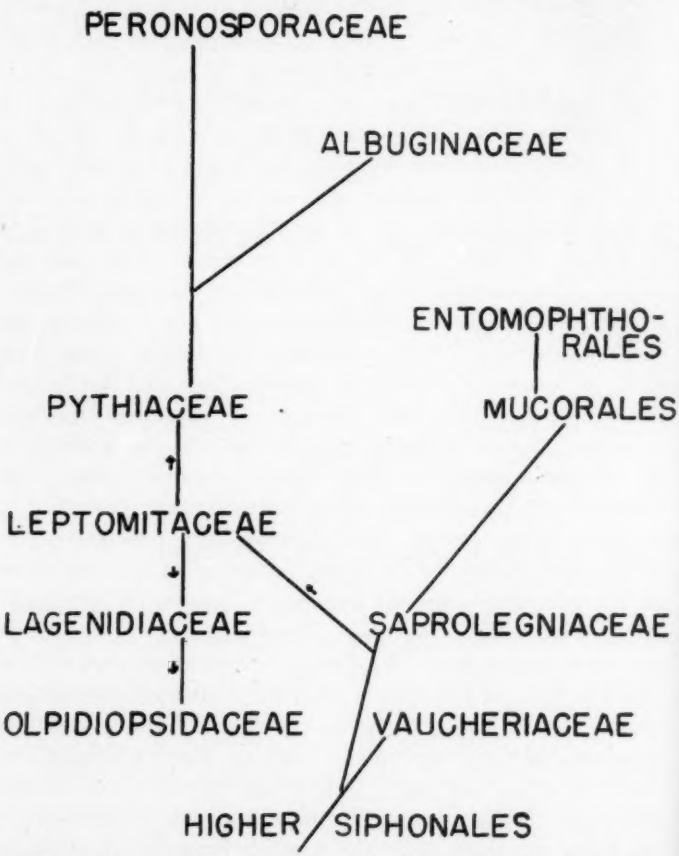

FIG. 2. Phylogeny of the biflagellate Phycomycetes, based upon Sachs' and Mez's idea of the origin of the Saprolegniales from the Siphonales.

cellulose reaction upon the application of chloriodide of zinc solution while those producing swarm cells with a single posterior flagellum usually do not respond to this test. *Olpidium radicale*, according to Schwartz and Cook (**26**), and the walls of the spo-

rangial sorus of some of the Synchytriaceae, according to Scherffel (**25**), are an exception and do show the cellulose reaction.

There are no clearly marked differences in the modes of sexual reproduction that can be definitely correlated with these swarm cell differences. In some genera, *e.g. Synchytrium* and *Olpidium*, equal swarm spores may unite by two's, the resulting zygote producing upon growth and development a thick walled sporangium or sorus of sporangia. For *Pseudolpidiopsis* and *Olpidiopsis* the method of sexual reproduction has already been described. In *Polyphagus euglenae*, in *Rhizophidium graminis*, according to Ledingham (**15**), and in *Diplophlyctis intestina*, according to Sparrow (**30**), the gamete nucleus is carried through a rhizoid to another organism of the same species.

Historically the Chytridiales in the broader sense have been treated very variously by the different students of the classification and phylogeny of fungi. Sachs (**23**) did not mention the group in the 1874 edition of his Lehrbuch but in 1882, in Vines' translation (**24**) they are grouped with the Protococcoideae in line with Sachs' idea that the fungi represented side-shoots, that had lost their chlorophyll, from algae at different levels of evolution. In 1884 Anton de Bary (**3**) suggested that they might have arisen by degeneration or simplification of Mucorineae or Ancylistineae, to use his names, down through the Rhizidiae and Olpidiae to the Synchytriae, or possibly that they had evolved by the loss of chlorophyll from unicellular algae, *e.g. Characium, Chlorochytrium*, etc., of the Protococcaceae. In the latter case if the Chytridineae are monophyletic the Mucorineae and Ancylistineae were probably descended from them. Possibly, he suggested, the Rhizidiae descended from these higher groups and the Olpidiae and Synchytriae came from the unicellular algae.

In 1901 Dangeard (**9**) suggested that the "Chytridinées" were descended from zoöspore-producing "Monadinées à nutrition animale" and that these are connected with *Vampyrella*. The latter is usually placed in the Rhizopoda. He believed that the chlorophyllless Protozoa, especially various groups of the Flagellata, are the source of several lines of evolution leading to plants by the acquisition of chlorophyll and the loss of the animal type of nutrition. The possibility of evolution in the opposite direction from

the lower plants to the one-celled animals he rejects as totally impossible. Nearly forty years ago Charles E. Bessey (**4**) strongly influenced by the ideas of Sachs, suggested that those organisms of this group which live internally in their host cells were derived from unicellular green algae, the Protococcoideae. On the other hand the "Chytridiaceae," that live externally on the host cell, obtaining their nourishment by rhizoids, he suggested might be derived from the neighborhood of *Botrydium* (one of the heterocont algae or Xanthophyceae). With our increased knowledge of the Chytrids it is clear that these suggestions need to be greatly revised, if accepted at all.

Atkinson (**1**) in 1909 wavered between the possibility that the Chytridiales had arisen from Protococcales that had lost their chlorophyll or that Dangeard's suggestion of their origin from the Monads might be correct. His studies of *Rhodochytrium,* which he considered an alga that rather recently had undergone loss of chlorophyll, made him not unfavorable to the idea of the algal origin of the whole Chytrid group. He, at that time, did not believe that the number or positions of the flagella on the swarm cells were of fundamental phylogenetic importance.

Mez (**20**), in 1929, in his discussion of the relationships of the fungi, as determined principally by the serum reaction method, made extrapolations for the Chytridiales, and other possibly related organisms which are too small or difficultly obtainable in sufficient quantity to permit of study by that method. He suggested that by parallel lines of reduction the Ancylistidaceae and Lagenidiaceae had arisen from the Saprolegniaceae and that from the Lagenidiaceae by further reduction had developed the Woroninaceae (Olpidiopsidaceae). The similarity in structure of these latter to the Olpidiaceae he considered to be due to convergence, the single posterior flagellum of the latter militating against the idea that they are related to the Woroninaceae with two anterior flagella. The Oöchytriaceae, Cladochytriaceae and Rhizidiaceae and thence the Olpidiaceae, he considered as a progressively simplified series arising in the neighborhood of the Monoblepharidales. This latter group he believed to have arisen, together with the Saprolegniales, from the Siphonales, the Saprolegniales from near *Vaucheria* and the Monoblepharidales from further down the line,

not far from *Codium*. In the Saprolegniales the anteriorly biflagellate structure of the swarm cells is retained, one flagellum, with a consequent reversal of position on the cell, being charac-

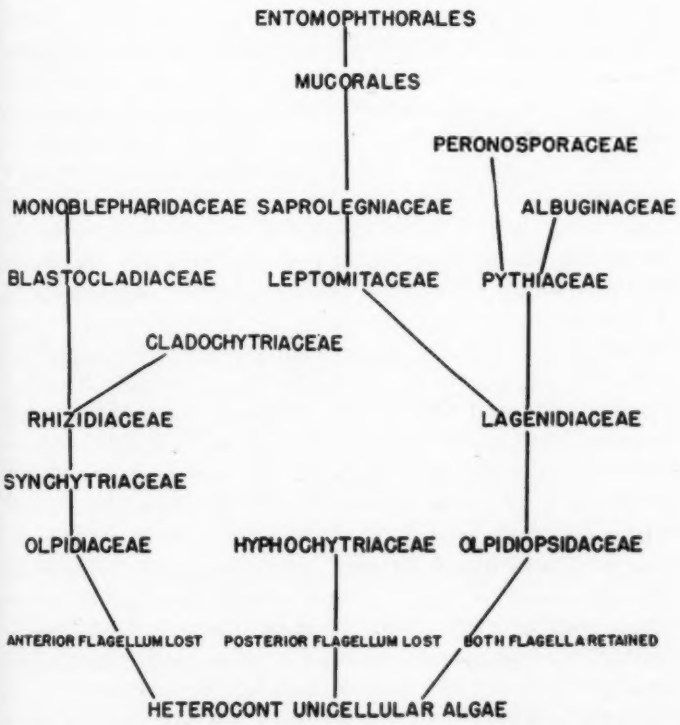

FIG. 3. Suggested phylogeny of the Phycomycetes based upon the idea of their origin from unicellular algae.

teristic of the Monoblepharidales. The serum precipitation reaction is very strong between the Saprolegniales and *Vaucheria*.

It seems pretty well agreed now (Sparrow (**29**), Weston (**33**), and others) that the posteriorly uniflagellate Phycomycetes form a continuous phylogenetic series. These would contain the four groups, which Sparrow breaks up into many families, of Olpidiaceae, Synchytriaceae, Rhizidiaceae and Cladochytriaceae, which for

Fitzpatrick made up the bulk of the Chytridiales, and the Monoblepharidales as delimited by Sparrow in 1933. The first three are monocentric, *i.e.* from the swarm cell arises a uninucleate cell which enlarges, becomes multinucleate and then becomes directly a sporangium or, in Synchytriaceae, a sorus of sporangia. In the Cladochytriaceae the organism is monocentric at first but by the passage of a nucleus through a modified rhizoid (rhizomycelium of Karling) to an enlarged portion of the latter it becomes polycentric, each center becoming a sporangium or budding off a cell or cells which become sporangia. In the Monoblepharidales we have coenocytic organisms varying from the clavate *Blastocladiella* which bears at its apex a single sporangium or gametangium to the larger filamentous forms like *Blastocladia, Allomyces* and *Monoblepharis* which are coenocytes and respectively isoplanogametic, heteroplanogametic and reproducing by fertilization of a non-flagellate egg by a uniflagellate sperm. The egg in some species of *Monoblepharis* is more or less motile though lacking a flagellum. The main gap in the whole series is between the unicellular, uninucleate Rhizidiaceae and the multinucleate, coenocytic *Blastocladiella.* In the former practically the whole contents of the single cell, after nuclear division, followed by cleavage of the cytoplasm, become the mass of swarm cells while in the latter the multinucleate clavate plant body gives rise at its apex to a single sporangium (or gametangium) whose contents become the swarm cells. Perhaps a step toward the latter is seen in those Chytrids which leave a little bit of nucleated cytoplasm in the base of the sporangium, from which a new sporangium may be formed. Possibly the basal vesicle of *Phlyctochytrium* may have been the initial point for the evolution of the vegetative base from which arises the sporangium in *Blastocladiella.*

It is of course easy to conceive of a reduction from some more filamentous form of the isoplanogametic *Blastocladia* through *Blastocladiella* to the Rhizidiaceae and other posteriorly uniflagellate Chytridiales and in the other direction to the closely related, heteroplanogametic *Allomyces* and to *Monoblepharis.* We must then find an origin for *Blastocladia.* To derive *Monoblepharis* from the alga *Oedogonium* seems far-fetched. The latter is truly cellular, with uninucleate cells and the zoöspores and sperms are

stephanocont, *i.e.*, have a wreath of cilia near the anterior end. *Monoblepharis* is coenocytic, not cellular, and the zoöspores and sperms are posteriorly uniflagellate. The cell wall composition and structure are different also.

The groups with biflagellate swarm cells: Olpidiopsidaceae, Lagenidiaceae, Leptomitaceae, Saprolegniaceae, and the three

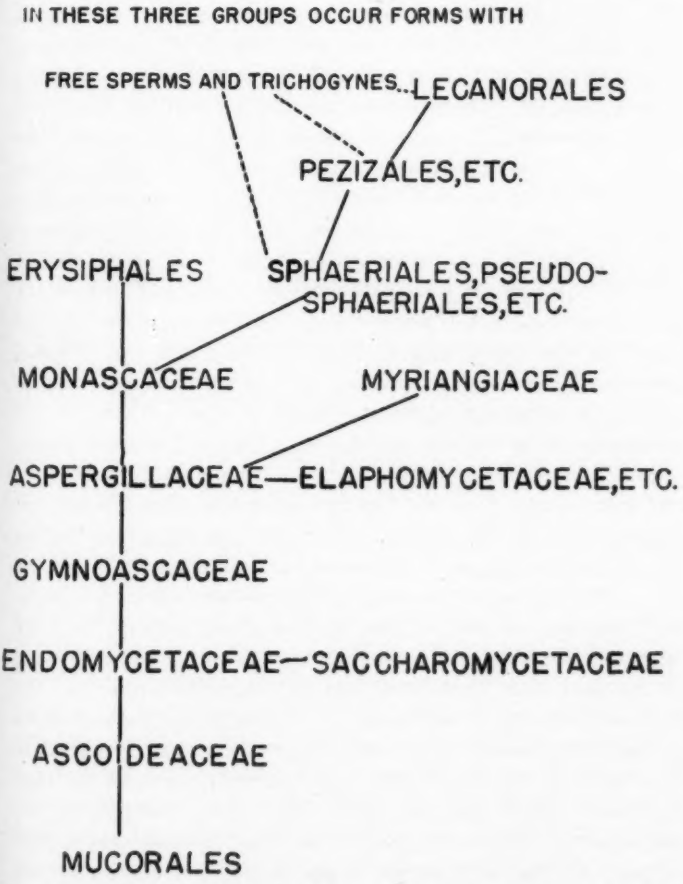

FIG. 4. Phylogeny of the Ascomycetes following in the main Dangeard, Atkinson, Gaümann and Mez.

families composing the Peronosporales also form a rather continuous series. In the first-named family the life history and general morphology are close to those of some of the Olpidiaceae but the swarm cells of the latter are posteriorly uniflagellate. Furthermore in the Olpidiopsidaceae the cell wall shows an immediate response characteristic of cellulose on application of the iodine-containing cellulose tests, while this reaction is slow or lacking in the *Olpidiaceae, Olpidium radicale* being an exception. Sexual reproduction in *Olpidiopsis* resembles that in *Pseudolpidiopsis,* being the union within the host cell of two adjacent uninucleate fungus cells, the contents of the one passing into the other, the latter after a longer or shorter resting period becoming the sporangium. In the Lagenidiaceae a short coenocytic mycelium, often limited to one but sometimes extending to several host cells, divides into short segments each of which becomes a sporangium, emptying by an exit tube, or a gametangium. The latter may be an oögone with a single egg, in some cases at least with periplasm, or an antherid whose contents enter the oögone through a conjugation tube. In the Leptomitaceae, Saprolegniaceae and the Peronosporales, the coenocytic mycelium is very much more extensive (except in some alga-inhabiting species of *Pythium*) and bears terminally on the main or lateral hyphae the sporangia and gametangia. This series of families is rather close, without great gaps, and may be read from the Olpidiopsidaceae upward through the Lagenidiaceae to the Leptomitaceae and Saprolegniaceae and to the Peronosporaceae. On the other hand the evidence is just as strong that the Lagenidiaceae are an intermediate step in the downward evolution to the Olpidiopsidaceae. Long ago Sachs (23) suggested that *Vaucheria* among the Siphonales was probably close to the algal form from which the Saprolegniales arose. If the observation of Apinis was correct that in the genus *Archilegnia* are produced flagellate sperm cells which enter the oögone through definite openings this shows still greater similarity between the alga mentioned and the Saprolegniales. Mez's serum reaction experiments reveal a very strong reaction between *Vaucheria* and *Saprolegnia.* The fact that the species of the genus *Woronina* are all parasites in the filaments of Saprolegniaceae except one, and that this species parasitizes in *Vaucharia* has been adduced as a further

argument for the close relationship of these groups. If this is accepted there was then a downwardly progressing simplification from the Saprolegniales through the Lagenidiaceae to the Olpidiopsidaceae and an upwardly progressing evolution to the Peronosporales with some *Vaucheria*-like alga as the starting point.

Vlk (**31**, **32**) has studied the structure of the flagella of the swarm cells of various algae and Protozoa and of one species of

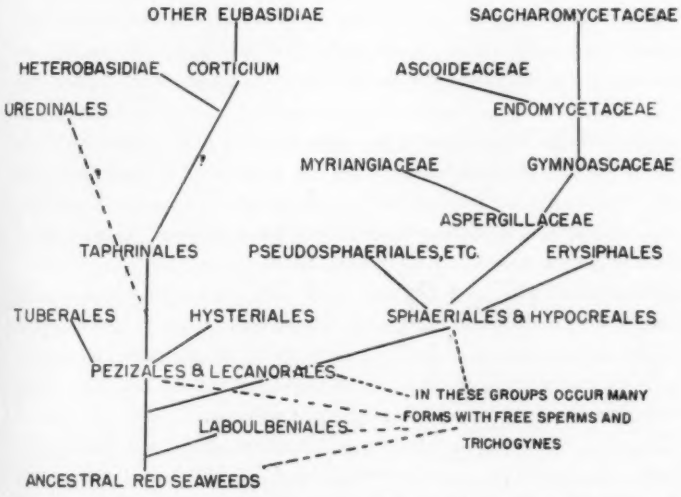

FIG. 5. Phylogeny of the Ascomycetes and origin of the Basidiomycetes based upon Sachs' idea of their derivation from Red Seaweeds.

Slime Mold, while Couch (**8**) has studied the flagellar structure for the three types of swarm cells in the Phycomyceteae, those with one posterior flagellum, with one anterior flagellum and with two anterior or lateral flagella. The latter has shown that for the biflagellate type, the series just under discussion, one flagellum, the anterior one, if they differ in position, is of the tinsel type and the other of the whip-lash type. Vlk has shown that in the three orders Volvocales, Protococcales and Ulotrichales the two or four flagella present on the swarm cells are of the whip-lash type only. The groups Chrysophyceae and Heterocontae (Xanthophyceae) produce one flagellum or two flagella on their motile cells,

the single flagellum (anterior in position) being of the tinsel type and the second, when present, of the whip-lash type and directed posteriorly. In the Euglenophyta the one or two flagella are of the tinsel type only, while in some of the colorless Flagellata the flagella, whether 1, 2, 4, 6 or 8, are all of the whip-lash type except in the Oicomonadaceae in which the single flagellum is of the tinsel type. Unfortunately for our speculations he has published no reports on the type of flagella in the Siphonales.

I am therefore tempted to suggest that from a group of one-celled Heterocont algae, by the loss of chlorophyll, as in some of the species of *Harpochytrium,* there arose a parasitic series, the Chytridiales in the broader use of the term, and that those that retained both flagella led to the Olpidiopsidaceae, those that lost the posterior, whip-lash flagellum led to *Rhizidiomyces* and its relatives, and that by loss of the anterior, tinsel-type flagellum arose the whole *Olpidium* to *Monoblepharis* series. If we could know whether the higher Siphonales have the two types of flagella (and some species of *Vaucheria* have unequal flagella) we would have greater faith that this group might well be the ancestral line of the Saprolegniales. If the two flagella should turn out to be both of the whip-lash type, as in the Ulotrichales, the alternate suggestion perhaps would have greater validity.

Figure 2 is a diagrammatic representation of the relationship in accordance with the ideas of Sachs and of Mez who derive the Saprolegniales from the Siphonales, near *Vaucheria.* In figure 3 is given a diagram showing how the Chytridiales might have arisen from unicellular algae.

The Mucorales have been tossed hither and yon by those seeking to connect them with lower Phycomyceteae. The fact that cellulose is concealed in many of them and that chitin seems to be present in the sporangiophores of some has led to the suggestion that they have arisen either from the Monoblepharidales (in the broader sense of the term) or from the Cladochytriaceae or their near relatives in which sexual reproduction is isogametic. I venture to suggest that they have arisen from soil Saprolegniales. In the first place the latter group contains many soil-inhabiting forms, as is true of the Mucorales. In the latter the young, rapidly growing mycelium often shows the presence of cellulose though the

cellulose reaction is usually masked by the presence of other substances in the older mycelium. In the Saprolegniaceae we find *Aplanes* in which the sporangium produces spores which lack flagella, a suggestion as to how the sporangia of the Mucorales may have arisen, with their non-motile spores. In *Dicranophora* the "zygospore" is the product of the union of a small, antherid-like gametangium with a large, oögone-like one. Even in those Mucorales where the gametangia are approximately equal in size the few cytological studies seem to show that the privileged nucleus (or nuclei) from one gametangium passes through a small opening into the other gametangium, after this occurring the complete dissolution of the septum separating the two.

Probably the Entomophthorales are close to the Mucorales, especially since the "conidia" of *Basidiobolus* have been shown by Miss Levisohn (**18**) to be sporangia which produce their spores only after having been ingested by frogs and other animals. Their sexual reproduction is not uniform but resembles somewhat that of various Mucorales. The Harpellaceae and Genistellaceae studied by Leger, Duboscq and Gauthier (**16, 17**) and the various genera of parasites of amoebae and nematodes studied by Drechsler (**10, 11**) and comprising the family Zoöpagaceae possess types of sexual reproduction that have some similarity to those found in the Mucorales and Entomophthorales, and possibly are related to them, but their exact relationship can probably be revealed only by life-history and cytological studies. Possibly they have developed in a direction somewhat parallel to these two orders from forms with slender filaments in the neighborhood of the Pythiaceae.

The higher fungi are still a battleground for the proponents of various phylogenetic speculations. The old Brefeldian (**6**) theory that sexuality was entirely lacking in them has been completely refuted. This is true also of his idea that the ascus was an asexual structure homologous to and descended from the sporangium of the Phycomycetes while the basidium was a conidiophore with fixed number of conidia (four, in the majority of the Basidiomyceteae) and descended from the conidiophore of Phycomycetes. It is now rather generally agreed that the ascus and basidium are homologous structures and that the latter has been evolved from

the former. Accordingly, any speculation as to the phylogeny of the one group must perforce have reference to the phylogeny of the other.

Brefeld looked upon *Monascus* as a fungus containing a single, large, multispored ascus, enclosed by a peridium of hyphal structure. This, he surmised, was the equivalent of a stalkless sporangium of *Morticrella,* surrounded completely by the envelope of hyphae usually found at the base of the stalk. The fact that the ascocarp of *Monascus* contains many asci formed in much the same way as those of *Aspergillus* throws *Monascus* out as an intermediate step from the Phycomyceteae to the Ascomyceteae.

Dangeard (**9**) and Atkinson (**2**), and following them many other mycologists have suggested that the Ascoideaceae, especially *Dipodascus,* might well be an intermediate stage between the Mucorales and higher groups of the Ascomyceteae. In *Mucor* the union of two multinucleate gametangia produces a multinucleate zygospore from which after a resting period there arises a sporangiophore bearing a large sporangium. Their suggestion is that there has been a telescoping of these two phenomena. In *Dipodascus albidus* the mycelium is composed of multinucleate (coenocytic) segments. From this mycelium, often from adjacent segments, arise two multinucleate gametangia which unite, with one privileged nucleus from each gametangium uniting to form a diploid nucleus. From the united gametangia without delay grows a long, usually tapering, multinucleate ascus within which are produced very numerous ascospores which are forced out of the apex of the ascus by the swelling of its contents or by the contraction of the stretched ascus wall, or both. From a primitive Ascomycete of this type it is suggested that by reduction in the number of nuclei per segment of mycelium the more typical monocaryon mycelium was derived. Indeed *Dipodascus uninucleatus* has that type of mycelium. Similarly the number of nuclei in the gametangium is assumed to have become reduced to one and the number of ascospores to have become standardized at eight or four. In this manner would have arisen the Endomycetaceae, the yeast-like derivatives of which are the Saccharomycetaceae. By a delay in the union of the gamete nuclei and their multiplication by conjugate division and then the branching of the zygote it is assumed that the

ascogenous hyphae arose, the nuclear union occurring in the terminal asci. The development of loose protective hyphae would give us the Gymnoascaceae and the transformation of these loose hyphae to a firm structure would produce the perithecial structures of the Aspergillaceae. From this it is then suggested that the Pyrenomycetes and eventually from them the Discomycetes have evolved. In all of these groups typically two gametangia unite, either equal in size, or modified into a smaller antherid and a larger oögone. Finally, at several points fertilization of an extension of the oögone by conidia instead of by antherids is assumed to have led to the production of spermagonia whose sperm cells fertilize the trichogynes in many Sphaeriales, Pezizales, Lecanorales and Laboulbeniales. This phylogenetic scheme is shown in figure 4.

Like many Mucorales the cell wall of the Ascomyceteae frequently contains chitin and only in rare instances, as in the ascogenous hyphae of some lichens, is cellulose so unmixed with chitin and other substances as to give the cellulose reaction when treated with chloriodide of zinc.

A further substantiation of the foregoing theory is the fact that the Mez (**20**) school claims a definite serum reaction between the "Protascales" and the Zygomycetes.

Guilliermond's (**13**) investigations on *Spermophthora* reveal an organism with an alternation of a coenocytic, branching gametophytic mycelium containing, presumably, haploid nuclei and a septate sporophytic mycelium of uninucleate cells with diploid nuclei. Terminal or subterminal gametangia are cut off by the formation of septa and within these are produced numerous fusiform gametes which are set free by the rupture of the gametangium. These unite, the nuclei uniting usually in the conjugation tube, and produce a few-celled, more or less branched mycelium of uninucleate cells, the terminal cells of which become spherical, eight-spored asci. He homologizes this limited monocaryon mycelium with diploid nuclei with the dicaryon ascogenous hyphae of more typical Ascomyceteae, the union of nuclei in the latter being deferred until the formation of the ascus. The union of distinct gametes is, in his opinion, a precursor of the condition in higher Ascomycetes where the two gametangia unite. This genus he considers as oc-

cupying an intermediate position between the Phycomycetes and Ascomycetes, with the Ascoideaceae, Endomycetaceae, etc. as lateral branches of the main line of ascent.

Anton de Bary, Atkinson, Gäumann, Mez and the great majority of modern mycologists held or hold to some form of the foregoing schemes, which derives the Ascomyceteae from the Phycomyceteae. Attention should, for the sake of fairness, be given to the hypothesis of Julius Sachs (**23, 24**) who suggested an entirely different mode of origin of the group. As one of the fundamental tenets of his theory of classification of the lower plants was the idea that fungi were polyphyletic and that those fungi that showed vegetative and reproductive structures similar to those of certain algae, are to be considered to have arisen from these algae by the loss of chlorophyll with the changes that such a loss would entail. Thus the Saprolegniales arose, according to him, from the Siphonales. The Ascomyceteae he suggests might have arisen from the vicinity of the red seaweeds. In the simpler members of this group, such as the Nemalionales, the plant body consists of branched filaments of uninucleate cells. Each transverse septum is centrally perforate, with a continuous cytoplasmic connection from cell to cell. Typically their sexual reproductive organs are an oögone with a long receptive extension, the trichogyne, and usually clustered antherids, often of the flask type, from whose interiors are pushed out the small, naked, sperm cells, with a large nucleus compared with the size of the cell. These non-motile sperms are transported by currents of water to the trichogynes to which they cling, secreting a thin cell wall and then dissolving an opening into the trichogyne into which the contents of the sperm pass. The sperm nucleus progresses toward the oögone, past the trichogyne nucleus which is present in a few forms, to the egg nucleus with which it unites. Usually with but little delay this zygote nucleus undergoes division which is meiotic in the majority of the Nemalionales or mitotic in a few of this order and in the other orders of the red seaweeds. These resulting nuclei divide further and pass out into short or long filaments (gonimoblasts) whose terminal cells singly or successively back from the apical cell, enlarge and become the carpospores. Each of these, it is apparent, contains a diploid nucleus, where the nuclear division in the oögone was

mitotic or a haploid nucleus, where that division was meiotic. In the majority of the whole class the carpospores give rise to plants more or less similar to the gametophyte, but with diploid nuclei, and producing tetrasporangia, instead of sexual organs, within which occur meiosis and the formation of naked tetraspores, which in their turn give rise to the gametophytes. The sporophytes are much reduced in some genera and in *Liagora tetrasporifera* Börgesen (**5**) showed that instead of carpospores, the terminal cells of the gonimoblasts are tetrasporangia.

The parallelism between red seaweeds and some of the Ascomyceteae is very striking: (1) Vegetative structures of filamentous, unicellular hyphae with perforate septa and continuity of cytoplasm from cell to cell, (2) formation in flask-shaped antherids of small, water-borne sperm cells, which are naked in the red seaweeds and in some of the Ascomyceteae, (3) presence of trichogynes on the oögones, to which the sperm cells cling and into which the sperm nuclei enter, (4) after the sperm nucleus reaches the oögone numerous branches arise from the latter into which pass the nuclei produced by the nuclear divisions in the oögone, (5) formation at the extremities of these branches of carpospores (in *Liagora* of tetrasporangia) in the red seaweeds, in the Ascomyceteae of asci in which meiosis occurs and ascospores are produced, (6) in some members of each group from the cells below the oögone arises a protective envelope surrounding the carpospores or asci, (7) several species of undoubted, higher red seaweeds lack chlorophyll and are parasitic upon other Florideae.

If the foregoing parallelism is accepted as indicating true relationship, and not an evolutionary convergence, it explains in a simple manner the occurrence in many Ascomyceteae of trichogynes and free sperm cells. The explanation of these by the advocates of the hypothesis of de Bary, Atkinson, Gäumann, etc. is that the primitive sexual reproduction in the class, was by the direct union of two gametangia and that by the progressive sterilization of the apical cells of the filament containing the female gametangium a trichogyne developed which could be fertilized by the male gametangium, and by further evolution, by conidia as substitutes for antherids, eventually these conidia becoming evolved into definite sperm cells. By the hypothesis of origin from the Florideae the

occurrence of the sperm cells and trichogynes is explained as a hold-over from the ancestral forms. *Collemodes* and *Ascobolus carbonarius* show the steps by which fertilization by free sperms changes to union of trichogyne with antherids. Progressive shortening of the trichogyne leads eventually to direct union of oögone and antherid.

If the Floridean ancestry of the Ascomyceteae is accepted the whole group must be stood on its head, as it were, as compared with the arrangement under the other hypothesis (FIG. 5). Those groups in which occur free sperm cells and oögones with trichogynes must be put first. Thus the Laboulbeniales with very Floridean types of sexual organs, many of the Pezizales and Lecanorales, and many of the Sphaeriales and Pyrenulales and some Hypocreales would occupy the more primitive position. It should be noted that it is among these fungi that the enveloping protective structures resemble most closely those of the red seaweeds. Instead of being the most primitive, the Aspergillales and Saccharomycetales would be the furthest evolved away from the primitive Ascomyceteae. The Taphrinales, especially if we include therein the Ascocorticiaceae, would be simplified forms of Pezizales with greater and greater disappearance of the excipulum and a more unlimited marginal growth of the hymenium.

As an argument against this hypothesis of the Floridean ancestry of the Ascomyceteae must be considered the failure of Mez to obtain by the serum diagnosis method any indication of the relationship of Ascomyceteae and Florideae. Furthermore the cell walls of the latter are composed of pectose-like substances or at best of cellulose-like substances. In the former cellulose and other carbohydrates are present in the cell walls but masked by the presence of chitin, with a very few exceptions.

It is accepted by most mycologists that the clamp connections of the Basidiomyceteae are homologous to the hooks or croziers of the ascogenous hyphae of the Ascomycetes. We must then postulate a common diverging origin of both classes from ancestors possessing these structures or the evolution of one class from the other. In origin and development the ascus and basidium are similar up to the final stage of spore-formation. We must therefore seek in the Ascomyceteae forms producing croziers on their as-

cogenous hyphae with other points of similarity that would suggest the possibility of evolution of one from the other. The Taphrinaceae have been suggested, but they lack the hook formation. The Ascocorticiaceae have been found by Rogers (22) to possess these. They are in general structure, except for the ascus, similar to *Corticium* or its relatives. Whatever source we choose for the origin of the Basidiomyceteae, we have to assume a mutation by which the internal ascospores became extruded into external pockets, thus forming the basidiospores. If the foregoing suggestion is accepted then the *Corticium*-like fungi represent the groups from which the rest of the class have developed. In some Basidiomyceteae the inner wall of the spore is free from the outer wall, suggesting that the latter is actually an extruded pocket containing a spore.

It must be pointed out that the well-developed spermagonia of the Uredinales and the development of receptive hyphae or trichogynes may indicate an origin further down in the Ascomyceteae than *Ascocorticium* in which these appear to be lacking. If that is the case, is the promycelium (or basidium) of the Uredinales and Ustilaginales, as well as the basidium of the Auriculariales closely related to the basidium of the Eubasidiae? Are the Tremellales and Dacryomycetales related closely to the Auriculariales? How about their relationship to the Thelephoraceae? Can it be that the three orders of "jelly fungi" are externally similar by convergent evolution only? Does the formation of sperm cells on the monocaryon mycelium of these orders and of some of the Eubasidiae indicate a relationship through the Ascomyceteae to the Florideae or are these structures merely analogous but not homologous to the sperm cells of the groups mentioned?

Whence came the Gasteromycetes? Are they evolved from the Agaricaceae by further and further development of angiocarpy? Or must we seek their origin from the vicinity of *Corticium* by the adoption of angiocarpy to produce simple forms like *Protogaster?*

It will probably be long before mycologists are in agreement with regard to the problems outlined in this paper. It is hoped that the suggestions given here may stimulate thought along these lines.

MICHIGAN STATE COLLEGE,
EAST LANSING, MICHIGAN

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THE MYCORRHIZA OF ZEUXINE STRATEUMATICA [1]

JOHN N. PORTER

(WITH 6 FIGURES)

Zeuxine strateumatica (L.) Schltr., an orchid native to southeastern Asia, was first reported growing in Florida in January, 1936, west of Fellsmere. Ames (2) has given a thorough account of its occurrence in Florida between that date and June, 1938, reporting that the plant was spreading rapidly throughout peninsular Florida and that, unlike other orchids, it was showing a close affinity for cultivated land, occurring in such places as lawns and drainage ditches. The problem which then presented itself was that of determining the factors which influenced the dissemination and successful establishment of the orchid in an entirely foreign locality. Most orchids, including the genus *Zeuxine*, are known to be dependent to some degree on mycorrhizal fungi; therefore, it was desirable to learn the identity of the fungus with which *Z. strateumatica* is living in symbiosis and also the conditions under which the germination of seeds and maturation could take place. Consequently, this investigation was undertaken in order to study the mycorrhizal relationships of the orchid in question.

DISTRIBUTION, GENERAL CHARACTERISTICS, AND MYCORRHIZAL RELATIONSHIP OF ZEUXINE

The members of the genus *Zeuxine* grow terrestrially in the rain forest at low altitudes in southeastern Asia and the neighboring islands. Previous to its discovery in Florida, *Zeuxine strateumatica* had been reported, according to Ames (1, pp. 276–277), from the Philippines, Afghanistan, India, Ceylon, Malay Peninsula, China, Assam, Japan, Java, and Amboina. Ames has propounded the interesting question of the origin of *Z. strateumatica*

[1] Contribution from the Laboratories of Cryptogamic Botany and the Farlow Herbarium, Harvard University, no. *194*.

in Florida and its ability to spread so rapidly once it was established. He considers the possibility to be very strong that protocorms of the orchid were introduced from China with seeds of the centipede grass, *Eremochloa ophiuroides* (Munro) Hack. The absence of previous records is a strong indication that this is not an orchid species which has hitherto had representatives in both hemispheres. The fact that it readily grows in cultivated areas precludes the fact that it might simply have grown unobserved in Florida until recently.

Zeuxine strateumatica is a plant which ordinarily grows as an underground stem, the latter being the center of an axis at one end of which are produced the roots and at the other, during the proper season, the leafy shoot and eventually the inflorescence, composed of small white flowers. The lower leaves are short and adhere rather closely to the axis but the more terminal ones are much longer, erect, and of a linear character. The plant, when mature, is usually six or more centimeters long. In Florida it flowers in January and soon produces a considerable number of seeds. By the first of April it has completely vanished from above the surface of the ground and is again pursuing life as a subterranean stem.

There is a dearth of published work on the mycorrhizal aspects of *Zeuxine,* which may be partially accounted for by the fact that members of the genus, inconspicuous in appearance, are of little horticultural interest. Burgeff (**4**) has found that within the genus *Zeuxine* itself there is exhibited a definite trend from the autotrophic condition toward that of saprophytism and that this trend is correlated with dependence upon a mycorrhizal fungus. Using certain species (*Zeuxine clandestina* Bl., *Zeuxine* sp., and *Zeuxine purpurascens* Bl.) as examples he observed that *Z. purpurascens* had less leaf area and green pigmentation than did the two preceding species but with this closer approach to saprophytism was associated a more uniform and thorough fungus infection.

Burgeff noted and described fungus infection in all three of these species of *Zeuxine* but succeeded in isolating the endophyte, *Rhizoctonia mucoroides* Bern., in only one case, namely, from an undetermined species of *Zeuxine* from Tjibodas, Java. Burgeff has also isolated *R. mucoroides* from the following genera of

orchids: *Phalaenopsis* and *Vanda* of the Sarcanthinae group of Pfitzer; *Trichopilea, Odontoglossum,* and *Miltonia* of the Oncidiinae; and *Goodyera, Macodes, Vrijdagzynea, Hetaeria,* and *Cystopus* of the Physurinae, to which group *Zeuxine* also belongs.

Bernard (**3**), who originally described the fungus, had isolated it from *Phalaenopsis amabilis* Lindl. and from *Vanda tricolor* Hook. He described the fungus as one which produces long aerial hyphae raised above the loose mycelial covering of the substrate. It forms on rich nutrient media a brownish-gray felt, the young cultures according to him being reminiscent of *Sporodinia* or *Mucor.* Branched monilioid chains of conidia anastomose to form small, irregular sclerotia abundantly on the surface of the substrate. These sclerotia are whitish at first but soon assume a deep brown color. Burgeff's (**5**) observations, however, did not completely coincide with those of Bernard. Burgeff found that very large and strong sclerotia were frequently formed and that there were different strains of the fungus. The strains from the terrestrial orchids related to *Zeuxine* were found to exhibit distinct physiological differences from the *Vanda* and *Phalaenopsis* fungi. The fungus in question is associated with orchids which taxonomically are very close together in the individual groups, although the groups themselves are well separated in the family Orchidaceae. These relationships indicate that there is a close and specific relation between orchid and fungus, and, based on the six genera of the Physurinae from which *R. mucoroides* has already been isolated, the suggestion may be made that this fungus will be isolated from other members of the group. The question now arises as to whether the isolates obtained by Burgeff are strains or are actually different species of *Rhizoctonia.* Unfortunately, as yet not enough is known about these fungi to determine this point with accuracy.

THE ISOLATION OF RHIZOCTONIA MUCOROIDES FROM ZEUXINE STRATEUMATICA AND DESCRIPTION OF THE FUNGUS IN CULTURE

Fungus isolations were attempted in February, 1937, from plants shipped by air mail from Florida. The roots were kept fresh by retaining around them a moist clump of the earth in which they

had been growing. Rhizomes as well as roots were thoroughly scrubbed with brush, soap and water and were sectioned after first being sterilized on the exterior with 7 per cent calcium hypochlorite. Since there is only a simple epidermis and not a velamen present in the case of *Zeuxine* the roots were merely cut into sections about 5 mm. long and inserted in the agar. The isolation medium was the "Mn + N" medium of Burgeff (**5**), consisting of the necessary mineral salts and three grams per liter of starch as a source of carbohydrate.

The first attempted isolation ended in failure, most of the petri plates remaining sterile and the others showing only a species of *Fusarium*. Since the portions to be placed in agar had been left in the sterilizing solution for an excessive amount of time (over one-half hour) it is to be supposed that the endophyte was prevented from growing, as were most of the contaminants. The absence of a velamen may also have been a factor contributing to the rapid penetration of the calcium hypochlorite into the cells which contained the mycorrhizal fungus. A second attempt was made in January of the following year from plants obtained from Florida in the same manner as described above. Care was taken this time that the roots and rhizomes be left in the calcium hypochlorite solution for only fifteen or twenty minutes. Considerable contamination occurred in these second cultures, but a radially growing fungus occurred quite uniformly and soon began to produce monilioid chains of conidia in abundance (FIG. 1).

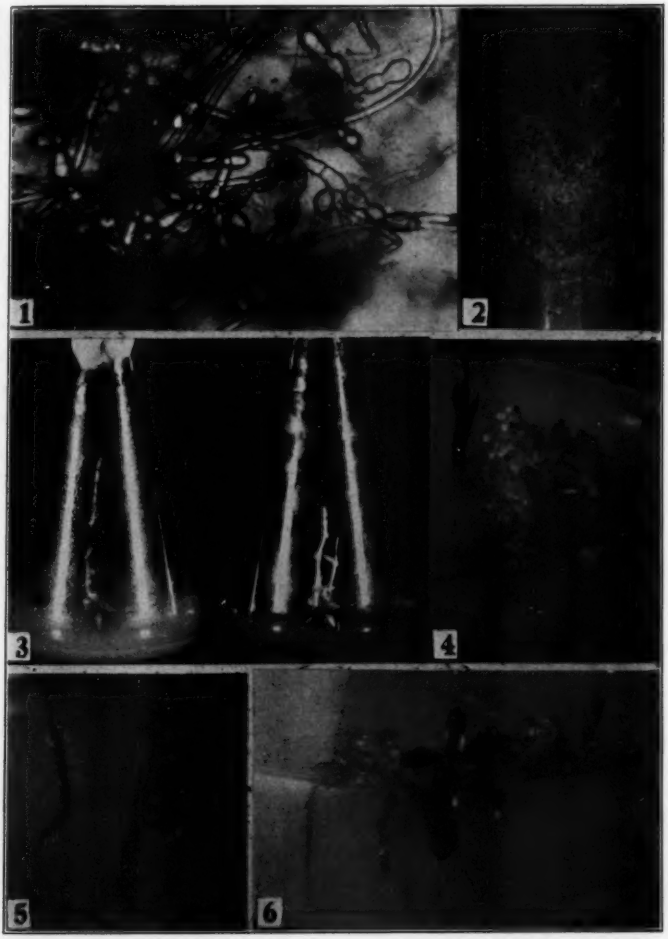
When grown in pure culture this fungus produced a light, cottony mycelium the color of which soon turned to dark brown. The hyphae showed some tendency to grow upon the glass sides of tubes or petri plates. Conidial chains of the type characteristic of the genus *Rhizoctonia* were abundantly produced in the agar, no other types of reproduction being observed, although brown sclerotia were formed from masses of monilioid conidial chains (FIG. 2). Measurements made at the end of a growth period of two weeks on Burgeff's "Of" stock medium (containing more starch than the "Mn + N" medium and no source of nitrogen) showed that individual cells of the chains of conidia averaged 21 μ long and 15 μ wide, while the hyphae averaged 6.4 μ in width. The figures just given compare very closely with Burgeff's figures

for *Rhizoctonia mucoroides* isolated from *Zeuxine* and other genera of the Physurinae. This fact, coupled with the close similarity of the general descriptions of the two fungi, leaves no doubt that *R. mucoroides* is the endophyte associated with *Zeuxine strateumatica.*

THE RESULTS OF ATTEMPTED SEED GERMINATION ON CULTURES OF THE ENDOPHYTIC FUNGUS

It is generally considered that the final test as to whether a given fungus is active in orchid mycorrhiza or is merely a soil saprophyte is its ability to promote germination of the seeds of the orchid genus from which it was isolated. Therefore, in order to determine whether the fungus isolated as described above would aid in germination of *Zeuxine strateumatica,* a quantity of seeds was obtained from Professor Oakes Ames in Ormond, Florida, soon after the fungus isolations were made. After being sterilized for 20–30 minutes in a filtered 7.5 per cent solution of calcium hypochlorite the seeds were sown February 14, 1938, on cultures of *R. mucoroides* growing on agar in 500 c.c. Erlenmeyer flasks. The medium used was the "Sb" medium of Burgeff in which starch is again the source of carbohydrate. Ammonium sulphate was used in place of the recommended sodium nucleinate, however, because it was more readily available. Six flasks were thus prepared, three of which were placed in diffuse light in the greenhouse and three in complete darkness, but at the same temperature (72° F. in winter). A small amount of additional seed material was obtained from the same source a short time later and these seeds were sown in three more flasks, each of which contained Knudson's (*8*) medium (with sucrose as a carbohydrate source) but no fungus. These flasks were placed in diffuse greenhouse light.

No evidence whatsoever of germination was observed by June 1, 1938, and further observations were not made until October 9 of the same year. Then it was noted that of the three flasks placed in the darkness two showed no germination, while in the third were two etiolated plants in close proximity, one 33 mm. tall and the other 46 mm. (FIG. 3). Of the three flasks originally placed in



FIGS. 1–6. 1, Mycelium and monilioid spore chains of *Rhizoctonia mucoroides* (× 240). 2, Appearance of *R. mucoroides* in stock culture. Sclerotia are being formed on the wall of the test tube (× ¾). 3, *Zeuxine strateumatica* plants one year after sowing. Flask on left has been kept in the light, that on right in the dark (× ⅓). 4, New shoot in originally illuminated flask at end of second year (× 1). 5. The same viewed from the bottom of the flask (× 1). 6, New shoot in flask originally placed in the dark; end of second year (× 1).

diffuse light all showed germination to some extent. In the second flask were counted nineteen seedlings, all 5–15 mm. long, but likewise not appearing above the surface of the agar and most of them occurring at its extreme edge. In the third flask there was only one plant (FIG. 3) but strangely enough it had attained a height of 74 mm., approaching the normal height of plants of this species in nature. The fact that almost all of the seedlings grew at the edge of the agar would indicate their need for the increased aeration or moisture occurring there. No germination whatsoever occurred with the seeds sown asymbiotically on Knudson's medium.

After the time of the examination just discussed, all flasks were kept in diffuse light in the greenhouse. The subsequent behavior of the seedlings was somewhat disappointing. It was hoped that at least one of the three large plants would produce an inflorescence by the first of March. However, they all collapsed upon the surface of the agar by mid-March and from then until the following autumn continued their existence as rhizomes below the surface of the substrate. Early in the fall of 1939 these plants had again produced shoots (FIGS. 4, 5, and 6) but this time they were much shorter (10–20 mm.) than those of the preceding year. No increase in height was noted after the middle of October, and their behavior in March was similar to that of the previous year. The many seedlings which were produced and then stopped growth beneath the surface of the agar remained quiescent, but alive, since their initial growth in the summer of 1938. On the other hand, the underground stems of the large plants gradually increased in diameter and length and obtained a certain amount of chlorophyll.

THE HISTOLOGICAL RELATIONSHIP BETWEEN ENDOPHYTE AND ORCHID

In order to study the position of the fungus within the tissues of the more or less dormant rhizomes, some of them were removed from the agar, cut into sections about five mm. in length, fixed in F.A.A. (formalin-acetic-alcohol), and stained by Durand's (7) method. The rhizomes, 15–20 cells in diameter, were found to be rather abundantly infected with the hyphae of *Rhizoctonia mucoroides*. Any cells, other than the epidermal cells or those of

the very slender central strand, were liable to contain fungus hyphae; infection, however, was limited to the cells of the side of the rhizome which was turned toward the agar. This, of course, applied to those plants which were growing at the extreme edge of the agar and between it and the glass. Cells were not observed in which digestion of the hyphae seemed to be taking place, all hyphae appearing to be in good condition. The nuclei of infected cells had lost their globular shape and were enlarged and distorted. The latter probably occurred as the result of the pressure of the massed hyphae on the nucleus. Those cells of the cortex which contained no fungus hyphae were partially filled with globular masses of starch. Such starch masses were completely lacking in infected cells.

DISCUSSION

That *Rhizoctonia mucoroides* is the endotrophic fungal symbiont associated with *Zeuxine strateumatica* in Florida is clearly indicated by the results of this investigation. Since the fungus seems necessary for the germination of seeds of this orchid and for its normal development and since the plant has begun to spread extremely rapidly in Florida one is led to suppose that *R. mucoroides* had already been established as a saprophyte in that area or as the symbiont of other species of orchids. The writer has been unable to discover any previous record of the isolation of *R. mucoroides* in America. Curtis (**6**), who has isolated from American orchids most of the mycorrhizal species of *Rhizoctonia* hitherto described, has not reported finding *R. mucoroides*. The most probable assumption of the origin of the fungus in Florida is that it had already infected protocorms of *Z. strateumatica* when they became mixed with seeds of centipede grass, and that these protocorms were a source of inoculum for the soil immediately surrounding the places where they were sown upon their arrival in Florida. The interval between the introduction of centipede grass into Florida (1917) and the period when *Z. strateumatica* became prevalent there may well be accounted for by the amount of time necessary for *R. mucoroides* to become established in the soil. A second possibility is that spore chains or sclerotia of the fungus

adhered to the grass and orchid seeds and were thus introduced at the same time. The sclerotia in particular would be able to withstand long periods of desiccation or other unfavorable conditions.

The fact that *R. mucoroides* has now been isolated from species of the genus *Zeuxine* in both Java and Florida is another important bit of evidence that there is an extremely close and specific relationship between orchids and their endotrophic symbiotic partners. That is to say, a single fungus species or a limited number of species are constantly associated with any given orchid species in nature. This fact in itself may account for the extreme geographical limitation of many species of orchids.

The synthesis of seeds with fungus has presented results which are interesting but not readily explanable. As previously stated, a need for the increased aeration and moisture occurring at the edge of the agar is probably the reason that most of the seedlings produced in the germination tests grew there. No valid explanation can be offered immediately concerning the three plants which obtained nearly normal size while the rest failed to push up from the agar. That the most rapid growth took place during the hot summer months indicates, however, that these plants are affected by temperature to a considerable extent. Temperature may also be an important factor in the failure of the one plant that reached normal size to produce an inflorescence.

Another possible answer to the perplexing question raised by the apparently aberrant behaviour of most of the seedlings has been supplied by the histological examination of the tissues. The absence of cells in which the digestion of fungus hyphae is taking place may explain the fact that the majority of plants produced have for a long time maintained a static rhizomatous condition in the agar. Normally, one finds cells in which digestion is taking place interspersed with those in which the hyphae are maintained in their normal manner (*Verdauungzellen* and *Pilzwirtzellen*), or these cells may be arranged in definite layers. It is considered probable by most workers in the field that the higher plant derives proteinaceous material and fatty substances from the fungus. If the usual digestion fails to occur one would then expect the higher plant to be arrested in development or parasitized. The lack of

proper aeration or moisture in the agar medium undoubtedly play their part in the inhibited development of these plants and may be the reason why the plants are unable to control completely their symbiotic partner.

The present investigation has been limited in scope since only a small amount of *Zeuxine* seed material has been available. However, because it develops very rapidly into a mature plant, *Zeuxine strateumatica* should be a satisfactory subject for future experimentation on orchid mycorrhiza. In particular, it would be very desirable to discover whether other species of *Rhizoctonia* isolated from various orchids not related to *Zeuxine* will promote germination in the latter.

The writer takes pleasure in expressing his appreciation to Professor William H. Weston, under whose supervision this study was made, and to Professor Oakes Ames for so kindly furnishing the seed and root material.

SUMMARY

1. *Rhizoctonia mucoroides* has been shown to be the mycorrhizal associate of *Zeuxine strateumatica* in Florida.

2. Seeds of *Zeuxine strateumatica* sown on cultures of *Rhizoctonia mucoroides* growing on the "Sb" medium in 500 c.c. flasks germinated in 6–8 months from the time of sowing.

3. Better germination was obtained in flasks kept in diffuse light in the greenhouse than in those placed in the dark.

4. No germination was obtained asymbiotically.

5. All indications are that the relation of fungus to orchid is, in this case, a specific one.

6. Because of its rapid development, *Zeuxine strateumatica* is worthy of future experimentation in the field of orchid mycorrhiza.

LABORATORIES OF CRYPTOGAMIC BOTANY,
HARVARD UNIVERSITY,
CAMBRIDGE, MASS.

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SOME SPECIES OF PAPULASPORA ASSOCIATED WITH ROTS OF GLADIOLUS BULBS [1]

H. H. Hotson [2]

(WITH 2 FIGURES)

The genus *Papulaspora* was erected by Preuss (2), in 1851, for an imperfect fungus which produced bulbils. Intervening work was thoroughly covered in 1912 by J. W. Hotson (2) in the introduction to his comprehensive monograph of the genus. This monograph and its 1917 revision (3) are the classic work on the genus with complete keys and detailed descriptions of the 25 species then known, and should be consulted for the taxonomy, morphology, and development. As is well shown in this monograph, these fungi are typically saprophytic in habitat and are found on dung, in the soil, and on various kinds of decayed fruits, etc. However, since B. O. Dodge and T. Laskaris (1) recently have found a *Papulaspora* associated with the rotting of *Gladiolus* bulbs, it seemed to the writer of interest to determine whether this species is indeed an active parasite on *Gladiolus* or merely a saprophyte, and, if a saprophyte, whether, perhaps, other species may not be involved. Toward this objective the following paper is merely a preliminary report, since the present emergency cut short the comprehensive study originally planned.

bulbs originally from Long Island, New York, kindly supplied, together with certain cultures, by B. O. Dodge to whom the writer is deeply indebted. From this material three species of *Papulaspora* were isolated, *P. Gladioli* (= *P. Dodgei*), *P. coprophila,* and

[1] Contribution from the Laboratories of Cryptogamic Botany and the Farlow Herbarium, Harvard University, no. *197*.

[2] This work was begun at the University of Washington under the guidance of J. W. Hotson; continued at Cornell University where facilities were offered by H. M. Fitzpatrick; and completed at Harvard University under the guidance and inspiration of Wm. H. Weston, Jr. To these men the writer is deeply indebted.

a third, which, as it proved to be new, is described as *P. appendicularis*. It is of interest to note that all of these were found under similar conditions as saprophytes on decayed bulb material resulting from previous infection by *Sclerotinia* (*Botrytis*) sp. The fourth species, *P. rubida*, previously found on *Gladiolus* in Pennsylvania by C. C. Wernham (**5**) and at first confused with *Urocystis Gladioli* (**4**) was very generously furnished by Dr. Wernham.

In making isolations, acid media, pH 4.8, were used, thus inhibiting bacterial growth sufficiently so that uncontaminated transfers to pure culture could be made. The fungi were cultured on potato-dextrose agar and on a synthetic medium whose formula, because it proved especially successful, is given here in the hope that it may be useful to others:

Starch (C.P.)	30 gms.	Peptone (Bacto)	5 gms.
Malt (Bacto)	10 gms.	Dextrose (C.P.)	10 gms.
Agar (Bacto)	15 gms.	Water (dist.)	1000 c.c.

Of the 25 species of *Papulaspora* which have been described, only one has been previously reported as such from *Gladiolus* bulbs, and this one and the other three have certain points in common. From the other species of *Papulaspora* these forms on *Gladiolus* may be separated by the following characters:

The primordium involves more than one cell; the mycelium lacks clamp-connections; the bulbils are yellowish-red to dark brown at maturity.

To facilitate the identification and differentiation of the species considered here, the following key will prove helpful:

A. Bulbils dark brown at maturity when seen in gross culture.
  B. Primordium a single lateral branch.
    C. Bulbils formed by the coiling of a single lateral branch in three dimensions, primordium not a spiral; conidia absent. 1. *P. Dodgei.*
    CC. Primordium a spirile, conidia borne on bottle-shaped sterigmata .....2. *P. coprophila.*
  BB. Primordium many lateral branches which fuse to form the bulbil; conidia borne on bottle-shaped sterigmata....3. *P. appendicularis.*
AA. Bulbils a brick red at maturity when seen in gross culture. 4. *P. rubida.*

In the following survey of these species, points of similarity and of difference will be considered in more detail.

Papulaspora Dodgei Conners, sp. nov.[1]

Papulaspora Gladioli H. H. Hotson.

A description of this species was provided by B. O. Dodge and T. Laskaris (**1**) and need not be repeated here. In a previous article, the writer (**4**) has shown that *Urocystis Gladioli* (Req.) Smith is distinct and separate from *Papulaspora Gladioli* (= *P. Dodgei*). This species is very striking in color, staining the medium dark brown and at maturity forming bulbils which are also dark brown. The staining of the culture medium is an easily recognizable character and one of the most striking. The bulbils average 44 μ in diameter with a range of from 24–64 μ. The growth of this organism is very rapid, mature bulbils being formed in 3–7 days. The majority of the writer's isolations from diseased bulbs were of this species and according to Dodge they were found in 20 per cent of the storage bulbs examined by him.

PAPULASPORA RUBIDA Hotson (J. W.)

This material came to Cornell from C. C. Wernham (**5**) as *Urocystis Gladioli* (Req.) Smith but upon further examination proved to be *Papulaspora rubida* which had been described by J. W. Hotson (**2**), in 1912. In the culture from Wernham, the fungus did not produce the spiral primordia which are characteristic of this species. However, comparison with the type material at the Farlow Herbarium leaves no doubt that these two fungi are identical.

Papulaspora appendicularis sp. nov.

Mycelium white, procumbent, profuse and matted; bulbils formed by the fusion of many lateral branches which usually arise around a central, septate hypha; more or less irregularly shaped, colorless when young, becoming light brown and finally dark brown at maturity; average diameter 60 μ with the extremes from 32–100 μ, often elongated and distinctly irregular (FIG. 1, $a + b$). Primordium not a spiral but consisting of many lateral branches. Conidia borne on bottle-shaped sterigmata especially in very young cultures (FIG. 1, c).

[1] See note at the end of this paper.

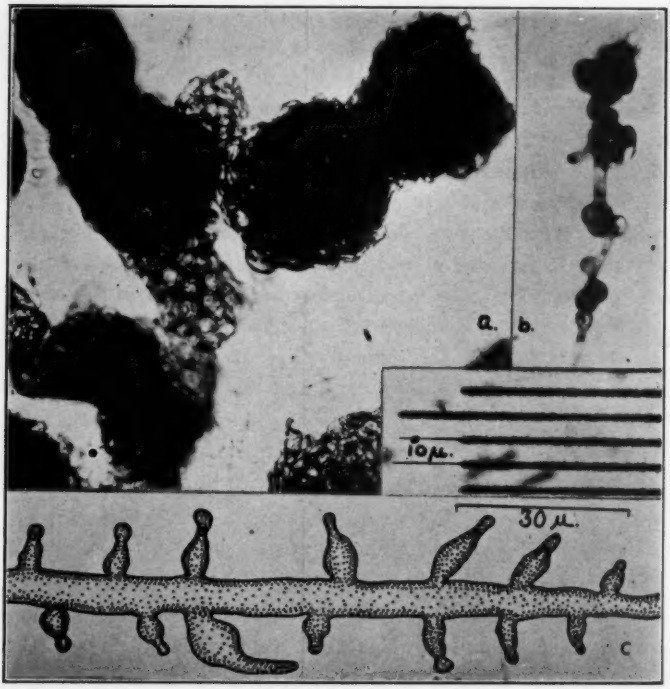

FIG. 1. *a* + *b*. Bulbils of *Papulaspora appendicularis* from prepared slides, × 400 ±, cf. scale. *a*, mature bulbils; *b*, early stages in bulbil formation; *c*, conidial stage, showing lateral bottle shaped sterigmata developing conidia, camera lucida drawing from living material, × 550 ±, cf. scale.

Mycelium album, procumbens; bulbilli e ramorum multorum lateralium, e hypha centrali septata ortorum, anastomosi efformata, forma elongati, irregulares, colore maturi atro-brunnei, 32–60–100 μ diam.; conidia in sterigmatibus lageniformibus producta.

This species, which appeared in very few isolations, is clearly distinguished from the others isolated from *Gladiolus* bulbs both by its many lateral branches and by the much larger size of its bulbils (FIG. 2). On comparison with other species of *Papulaspora* this species shows some general resemblance to *P. polyspora* Hotson (J. W.) but the latter has even larger bulbils (119–122 μ in diameter) composed of distinctly angular cells. After careful

comparisons the characteristics of the present species seem sufficiently distinctive to justify describing it here as a new species, *P. appendicularis.*

TYPE MATERIAL.

Farlow Herbarium, Harvard University.
Herb. of Plant Path. Dept., Cornell University.

PAPULASPORA COPROPHILA (Zukal) Hotson (J. W.)

Although this species, originally described by J. W. Hotson, in 1912, has been isolated from soil and has been reported as growing on dung, in this case it was found growing saprophytically on bulbs of *Gladiolus* previously attacked by *Sclerotinia* (*Botrytis*) sp.

The coiled primordia are most conspicuous in the younger stages of the growth of this species. In 50 hour cultures this character is easily seen as well as the conidia which are borne on bottle-shaped sterigmata. However, when this species is grown for a period of time on artificial media it loses its ability to produce these spirile primordia. Whether this fungus can be made to produce these structures again is an interesting problem which should be worked out. Possibly nutrition is involved and the artificial media lack certain ingredients essential for normal growth and spirile production by the fungus. The purpose of these conspicuous coils is not known. Whether they are vestigial sex organs which have degenerated or whether they are undeveloped sex organs is a question which has not, as yet, been answered.

The foregoing survey brings up certain points of interest. Within the limited area of Long Island, N. Y., there are besides *Papulaspora Dodgei* two other species which occur as saprophytes upon the diseased bulbs of *Gladiolus.* Of these, *Papulaspora coprophila* was described by J. W. Hotson, in 1912, but has never been previously reported from this source, while *Papulaspora appendicularis* is a new species as yet reported from this source alone. In addition, *Papulaspora rubida* has been found on *Gladiolus* in Pennsylvania. By examining material from other sources it may be possible to discover additional *Papulasporas* associated with the rots of *Gladiolus* bulbs. It also is possible that there may be some

species of *Papulaspora* associated with rots of *Gladiolus* bulbs in Europe, which because of the confusion with *Urocystis Gladioli* (W. G.) Smith have been overlooked. In any case, this survey, although limited, seems to indicate that there is here an ample field for future work.

In conjunction with the foregoing it seemed of interest to the writer to ascertain whether *Papulaspora Dodgei,* the only one of the species on this host which has been suspected of parasitism,

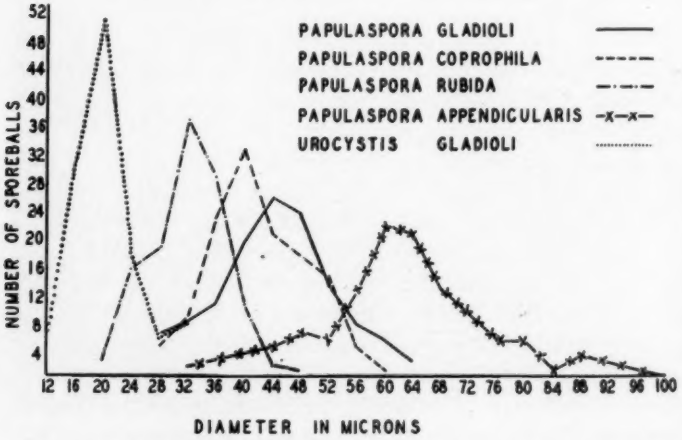

FIG. 2. Graph showing comparative sizes of bulbils of four species of *Papulaspora* from *Gladiolus* and also the spore-ball sizes of *Urocystis Gladioli.* Based on 200 random measurements of each.

really causes a disease. Since former workers were unable to reinfect the *Gladiolus* with this organism and because some doubt has been cast on the pathogenicity of this fungus, the writer carried out the following experiments.

In the greenhouse, 48 plants were used; 24 at 70° F. and 24 at 80° F. Eight of each of the three varieties, Alice Tiplady, Souvenir, and Halley were used, four being controls and four being inoculated in each series. The young shoots were inoculated from culture when they were about one inch in height and kept under normal growing conditions. Two of the four were inoculated by wounding and two by inserting the bulbils of the *Papula-*

spora inside the leaf sheath. The tests were run for a period of three months and were apparently completely negative since all the inoculated plants as well as the controls remained healthy.

In addition to this, it seemed interesting to carry out similar inoculations on these bulbs grown in nutrient solutions, *in vitro,* rather than in soil. Accordingly, three of each of the varieties were grown in nutrient solutions (Hoagland's) in the laboratory and were inoculated with the fungus. Although they had to be potted and transferred to the greenhouse at the end of three weeks because of mold contaminations, yet after three months the results were apparently negative as both the inoculated plants and the controls remained healthy.

Hence, from the point of view of the grower, this *Papulaspora* at least is not important as a cause of bulb loss, for which some other organism, often *Sclerotinia* (*Botrytis*) sp., may be responsible. This seems to be corroborated by evidence from the writer's many isolations from *Gladiolus* for the several *Papulasporas* seemed to accompany the one regularly occurring *Botrytis* as a saprophytic secondary invader.

SUMMARY

Associated with the rots of the bulbs of *Gladiolus* the writer has isolated the following species: *Papulaspora Dodgei* Conners, *P. rubida* Hotson (J. W.), *P. appendicularis* Hotson (H. H.), and *P. coprophila* (Zukal) Hotson (J. W.). Of these *P. appendicularis* is here described as new. These fungi are saprophytes living on the decayed bulb tissue caused by the primary infection of some other organism, in this case, *Sclerotinia* (*Botrytis*) sp., and by themselves do not cause a disease of the *Gladiolus.*

BIOLOGICAL LABORATORIES,
HARVARD UNIVERSITY,
CAMBRIDGE, MASSACHUSETTS

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NOTE

Dr. I. L. Conners of the Central Experimental Farm, Ottawa, Canada, in a letter to the writer, has called attention to the fact that Hotson (Mycologia **34**: 52–58. 1942) had created a homonym in describing *Papulaspora Gladioli* Hotson, a name preëmpted by *Papulaspora Gladioli* (Req.) Dodge & Laskaris, and sent the writer a copy of the manuscript he intended to publish to correct the mistake. In view of the fact that Mr. Hotson is in the armed services, and also in view of the fact that Conners' manuscript did not completely cover the situation, and also to save delay in printing the present paper without having to perpetuate an error, the writer feels that it is desirable to clarify the nomenclatorial situation. There is little doubt that the material communicated by Miss E. M. Wakefield and cited in Hotson's earlier paper is *Urocystis Gladioli* W. G. Smith and accordingly that name is valid, but there is some doubt concerning the status of *Uredo Gladioli* Requien since the original description could apply to either *Urocystis* or *Papulaspora*. In the Curtis Herbarium, however, there is a specimen from Duby, labelled in Curtis' handwriting:

Uredo Gladioli Duby!
fol. Gladioli
Duby

It would seem that this is an authentic specimen, if not a part of the type, as would be implied by the exclamation point. An examination of this specimen shows it to be the telial stage of the rust, *Puccinia Gladioli* Cast., of which, according to the International Rules, Article 57, *Uredo Gladioli* Req. becomes a synonym, and, according to Article 54, paragraph 2, so does *Papulaspora Gladioli* (Req.) Dodge & Laskaris. Since *Papulaspora Gladioli*

Hotson is invalid because a later homonym, it becomes necessary to assign to the imperfect fungus a new name and consequently, since this was the name proposed by Conners in his intended paper, it is only fitting and proper that the species should be labelled **Papulaspora Dodgei** Conners sp. nov., and that *Papulaspora Gladioli* Hotson be considered a synonym thereof. The Latin diagnosis required to validate the species, follows:

Mycelium primum albidum, profusum; hyphis septatis, cellulis multinucleatis; bulbulis ex stipitibus septatis oriundis, pallide brunneis vel atro-brunneis, sphaericis, 29–64 μ diametro, cellulis centralibus 1–6 raro pluribus, atro-brunneis, multinucleatis, cum corio unico cellularum pallide fulvarum circumdatis; hyphis primordialibus prope apicem spiraliter convolutis. Conidia absunt.

DAVID H. LINDER
FARLOW HERBARIUM
CAMBRIDGE, MASS.

TWO CASES OF UNUSUAL DEVELOPMENT OF FRUIT BODIES[1]

CLYDE M. CHRISTENSEN

(WITH 3 FIGURES)

Those who are familiar with fleshy Agaricales know that the geotropic response which results in an orientation of pores, gills, or other spore bearing surfaces perpendicular to the surface of the earth, sometimes goes awry, especially if fruit bodies are diseased or mechanically injured. Fruit bodies produced in unnatural environments, as on agar cultures or on wood in jars, sometimes exhibit a capricious orientation of pores or gills, indicating that forces other than gravity are involved in such orientation. Following will be described two interesting cases of such nongeotropic development.

The first case involves an agaric (*Russula* sp., tentatively identified as *R. atropurpurea* Peck) observed at Itasca Park, Minnesota, in September, 1937. More than a decade before that time several pits about 5 feet square and 4 to 5 feet deep had been dug for experimental purposes in the level ground in a Jack pine stand. During a rainy spell in September, 1937, when fleshy fungi were rather abundant, the writer observed 2 fruit bodies of this species of *Russula* growing out of the vertical walls of one of the pits. One appeared about a foot from the surface of the ground, the second about 2 feet from the surface and on an adjoining wall. The stem of each, although short, extended straight out from the vertical wall, no upward curve being visible. The cap in each case was parellel to the wall. The gills were normal, although the free edges of those on the upper side bent over as they lost their turgidity with age, as can be seen in figure 1. In other words, the fruit bodies were oriented in the same way to the perpendicular surface from which they grew, as fruit bodies growing on approximately

[1] Paper No. *457*, Misc. Journal Series, Minnesota Agricultural Experiment Station.

FIG. 1. *Russula* sp. growing out of the vertical wall of a pit. The surface of the ground is indicated by the carpet of needles; needles also cover the floor of the pit in the lower right corner of the picture. FIG. 2. Two sphaeroid fruit bodies of *Daedalea confragosa* which developed from normal fruit bodies. FIG. 3. A portion of the elongate pores on the central part of the lower edge of the lower fruit body in figure 2 enlarged to show the smaller horizontal pores in the walls of the larger ones.

flat ground are oriented to that. Both fruit bodies were attached to mycelium in the side of the wall, and were observed in their development over a period of two or three days. Certainly gravity had very little influence on the growth of the fruit bodies.

Second case. In October, 1940, a trunk of a dead willow two to three inches in diameter, collected in a swamp near Minneapolis, and bearing nine fruit bodies of *Daedalia confragosa* (Bolt.) Fries in various stages of maturity, was brought into the laboratory. The pores of all these fruit bodies were mainly daedaloid, only a few being lamellate. Since the fruit bodies were quite fresh, and it was thought that they might continue to enlarge, the trunk was placed in an upright position in a quiet corner of the laboratory,

with the lower end in a jar containing about 6 inches of water. It remained in this position throughout the period of observation, approximately six weeks, the water being renewed as necessary. The pores of the two lower fruit bodies continued to elongate, and in several weeks had grown about a quarter of an inch in length. They retained their normal shape and size, but very numerous smaller pores developed horizontally through their walls (FIG. 3). At the same time, pores arose over most of the upper surface of these two fruit bodies, transforming them into globoid structures illustrated in figure 2. These pores on the upper side pointed outward from the center of the fruit bodies. As can be seen in the illustration, they are smaller and more regular in shape than the pores on the lower side of the fruit bodies.

The hymenium of these anomalous pores, both the small pores extending horizontally through the pores on the lower side of the fruit bodies and those on the upper side, appeared normal, bearing immature and mature basidia, upon which occasionally four sterigmata could be seen. Cystidia with branched tips also were present. Few basidiospores were found in any of the pores, but this may have been due to the relatively dry air of the laboratory.

None of the upper seven bodies continued to grow after the tree trunk was brought into the laboratory, doubtless because of lack of water. Since the position of the trunk in the laboratory during the period of abnormal growth was approximately the same as its previous position in the field, and since the force of gravity obviously was about the same in the two locations, forces other than gravity must have been involved in the orientation of the pores. Apparently the normal tropic response to gravity is conditioned by a rather delicate balance that can be upset easily.

These two cases present exceptions to the general rule of geotropic response in this group of fungi. While one obviously cannot doubt that the formation of fruit bodies of Agaricales normally appears to be influenced by gravity, it may be fairly questioned whether geotropic growth responses regulate the formation of such fruit bodies so much as we have supposed.

UNIVERSITY FARM,
UNIVERSITY OF MINNESOTA,
ST. PAUL, MINNESOTA

NEW PROPOSALS RELATING TO THE GENERA OF THE BOLETACEAE

WALTER H. SNELL

(WITH 1 FIGURE)

A short time ago (Snell, **1941**), a modification of Gilbert's arrangement of the genera of the Boletaceae (**1931**) was presented as an improvement upon the one most commonly used on this continent —the tri-generic scheme of Peck (**1889**), which used as tribes of the genus *Boletus* the subgroups of Fries (**1863** & **1874**) with minor modifications and additions. Recently, Rogers (**1941**) demonstrated that S. F. Gray's *Natural Arrangement of British Plants* (**1821**) is post-Friesian, at least as far as the first volume of the *Systema* is concerned. Accordingly, Gray's generic names must be given the consideration that has been denied them by recent mycologists in the uncertainty that has prevailed concerning the priority status of the *Natural Arrangement*.

In Gray's family Hymenotheceae, two of the subfamilies (or tribes?) are the Boletideae and the Suillideae. The Boletideae contain species that are present-day Polyporaceae. The laterally attached species, whether corky, coriaceous or woody, are placed in the genus *Boletus,* but obviously this use of the name is of no interest to us because in the first volume of the *Systema* a few months before, Fries had established that genus in the present-day Boletaceae. The Suillideae include the forms that are fleshy and have long tubes separable from the cap and either united or distinct from each other, in the four following genera: *Suillus* Micheli, *Pinuzza* Micheli,[1] *Leccinum* Micheli,[1] and *Fistulina* Persoon.

Fistulina is obviously of no concern in this discussion.

[1] Gray (p. 646) ascribes these two genera as well as *Suillus* to Micheli, but apparently erroneously so, unless other publications of Micheli's were available to him that are not known at the present day. In the *Nova Genera Plantarum,* there is no mention of such genera as *Pinuzza* and *Leccinum,* although the Italian words "pinuzzo" and "leccino" along with "porcino," "pinaccio," "pinarello" and many others are given as the common names of fleshy fungi ("leccino giallo" for an agaric apparently, and the others for "pinophilous boletes").

Suillus was to include those forms with collar (annulus) distinct and had one species, *luteus; Pinuzza* had a fibrous annulus and one species, *flava*. Since both of these species belong in the Viscipelles of Fries and Peck, or the genus *Ixocomus* Quélet (**1888**), both names have priority over *Ixocomus* and either might be selected for the species with viscid pileus, adnate to decurrent tubes and rather small, elliptical spores. It would seem that *Suillus* should be preferred because it has a tradition which *Pinuzza* lacks, even though it also has had a more varied history. So far as is known, the name *Pinuzza* was not used before Gray and otherwise is of very infrequent occurrence in the literature. In fact, it is very difficult to find the name in synonymy. On the other hand, *Suillus* is an ancient name. According to Pliny (77 A.D.), the Romans used it (not in a generic sense, of course) for what was apparently *Boletus edulis* and perhaps other *Boleti,* while calling *Amanita caesarea* by the name *Boletus*. Caesalpino (**1583**) and Porta (**1592**) used both words in the same manner. Micheli (**1729**) first used *Suillus* as a generic name, applying it to the *Boleti* and using *Boletus* as did Tournefort (**1694** & **1700**) for the morels and phalloids. He was followed by Haller (**1742**, and in part, **1768**), Müller (**1763**) and Adanson (**1763**); Vaillant (**1727**) and Battarra (**1755**) used *Boletus* but not *Suillus*.

Up to this point, a majority had referred to the *Boleti* under the name *Suillus,* the notable exceptions being: Tournefort, who used *Fungus* for at least a part, and his follower, Vaillant; Dillenius (**1719**), who first used *Boletus* for this group and some of the polypores; and Battarra, who coined a new name, *Ceriomyces*. It was Linnaeus (**1753**) who definitely turned the tide away from *Suillus,* for which he substituted *Boletus* in Dillenius' sense, just as he changed the senses of all the names used by the Romans. Linnaeus was followed in this usage by: Schaeffer (**1762–1774**); Scopoli (**1760** & **1772**); Jacquin (**1773–1778**); Batsch (**1783–1789**), who, however, used the subgroup Suilli for the boletes; Bulliard [2] (**1791–1812**); Schrader [2] (**1794**), Persoon (**1801**) and

[2] Fries (**1821**, p. 386) and Bataille (**1908**) both stated that Bulliard and Schrader interpreted *Boletus* as the *Boleti* of the present day. Both workers, however, included the polypores in this genus, although they separated them in different subgroups.

others. Poiret (**1806**) resurrected Micheli's name *Suillus*[3] for the *Boleti* and one polyporaceous species, *betulinus* (see last sentence of footnote 2). Then Gray, as noted above, restricted *Suillus* to one group of the Boleti included in Fries' Viscipelles of *Boletus,* only to have it ignored by subsequent workers until Karsten in 1882 applied it in a still different sense, to make a new genus for the Friesian Cariosi (*cyanescens, castaneus,* etc.), the later genus *Gyroporus* of Quélet (**1886**).

Leccinum, used by Gray for species with no annulus, was necessarily a conglomerate genus, including the species: *aurantiacum* (with varieties *leucopodium* and *rufum*) and *scabrum* of the Friesian and Peckian Versipelles and the more recent genera *Krombholzia, Krombholziella* and *Trachypus; lactifluum* (= *B. granulatus*) and *piperatum* of the Viscipelles and genus *Ixocomus; subtomentosum* of the Subtomentosi and genus *Xerocomus; constrictum* (= *B. cyanescens*) of the Cariosi and genus *Gyroporus; edule* and *elephantinum* (= variety of *edule*) of the Edules, and *luridum* and *rubeolarium* (= *luridum*) of the Luridi and genus *Boletus.* With the exception of *piperatum,* the foregoing is the order in which the

[3] It is not a matter of great importance, but the situation may as well be clarified, especially since in the treatment of the Boletaceae in *North American Flora* **9**: 154, 1910, the first synonym under the genus *Boletus* (Dill.) L. reads as follows: "*Suillus* Poir. in Lam. Encyc. **7**: 496, 1806." In the first volume of the *Encyclopédie Méthodique,* Lamarck's genus *Agaricus* was tubular and included both the boletes and the polypores. Then Poiret, who prepared the material for the later volumes, inserted *Suillus* apparently as a genus, only to confuse the issue in a statement to be found below. The two pertinent paragraphs are given verbatim herewith:

"SUILLE. *Suillus* Genre de plantes acotylédones, de la famille des champignons, qui renferme un certain nombre d'espèces, d'une substance ordinairement ferme & coriace, munie d'un pédicule qui soutient un chapeau, dont la surface inférieure est munie de pores nombreux, très-sérres, alongés, tubulés, adhérens ensemble, mais *faciles à détacher de la substance charnue qui leur sert de réceptacle.* Ce dernier caractère est le seul qui les distingue des *bolets* (*boletus* Linn.; *agaricus* Lam.), la masse des tubes ne pouvant être, dans ceux-ci, separée de la substance charnue.

"Il est aisé de reconnoitre que les suilles, d'après ce caractère, ne sont qu'une division des bolets, & qu'ils ne peuvent pas en être séparées comme genre. Nous ne les présentons ici que parce qu'ils nous offriront l'occasion de rappeler plusieurs espèces qui n'ont pas été mentionnées à l'article AGARIC, dénomination qui avoit été adoptée par Tournefort, & que M. Lamarck a substituée à celle de *bolet* Linn. Nous nous bornerons cependant aux espèces les plus remarquables."

species were presented by Gray. As far as can be ascertained, the genus *Leccinum* has been used by no one else.

As suggested three paragraphs above, it is proposed to adopt *Suillus* Micheli ex S. F. Gray [type species—*S. luteus* (L. ex Fr.) S. F. Gray] for the Viscipelles of *Boletus* of Fries and Peck in place of *Ixocomus* Quélet. *Suillus* has priority over every other name for this group of Boleti and it is legitimate under the International Rules to select it even though Gray placed species to be included in this genus in two other genera. Furthermore, the establishment of *Suillus* (**1821**) in place of *Ixocomus* (**1888**) will obviate the raising of embarrassing questions such as why *Cricunopus* or *Rostkovites* (both Karsten, **1881**) or *Viscipellis* Quélet (**1886**) should not supersede *Ixocomus* Quélet (**1888**), and also why *Suillus* Karsten (**1882**) in an entirely different sense (for the Cariosi of Fries and Peck) should not supersede *Gyroporus* Quélet (**1886**).

It is also proposed that *Leccinum* S. F. Gray [type species—*L. scabrum* (Bull. ex Fr.) S. F. Gray] be adopted for the Versipelles of Fries and Peck in place of *Trachypus* Bataille (**1908**). The name used for this group by many mycologists in Europe has been *Krombholzia* Karsten (**1881**). This name, however, was originally used by Ruprecht [4] (1842) for a member of the Festuceae of the Gramineae, and R. Maire (**1937**) in its place suggested *Krombholziella,* which, however, is antedated by *Trachypus.* To be sure, Gray had no such conception of the use of the name *Leccinum,* for in this genus he placed species now found in several of the newer genera, but it is proper arbitrarily to select the first two presented by Gray under that name. Furthermore, in view of the previous confusion and the as yet unaccomplished establishment of the genus *Trachypus* because of its recent proposal (*cf.* Snell, *loc. cit.,* **1941**), a new name is not going to be greatly disturbing.

Since Gray used generic characters unacceptable today and accordingly made generic groupings which no one now would consider at all satisfactory, and especially since his genus *Leccinum* is so heterogeneous in the light of present-day understanding of its species, it may be objected that the use of any of Gray's boletaceous

[4] *Krombholzia* Rupr. ex Gal. in Bull. Acad. Brux. ix. II. 247 (1842), *nomen nudum;* ex Fournier in Bull. Soc. Bot. Belg. xv. 464 (1876).

genera is merely adding unnecessary confusion. On the other hand, these are times of change in mycology because of the discovery of new facts and the development of new taxonomic concepts, and one may as well unflinchingly make such changes as are necessary or desirable under the Rules and make them once and for all. Certainly, under the present Rules any changes made to Gray's genera are not going to be superseded, unless by special rulings. Further, in the Boletaceae, none of the more recent generic revisions has as yet been generally adopted in Europe, and in this country no revised scheme has had any vogue. Accordingly, a few generic changes at this time are not going to prove very disconcerting; it is believed that the suggested changes will lay more difficulties than they raise.

It is further proposed that *Versipellis* Quélet (**1886**), which antedates *Xerocomus* Quélet (**1888**), be considered a *nomen ambiguum*. All who have followed Fries and Peck and some who have modified the Friesian and Peckian schemes have used the term Versipelles for a tribe or group which includes the common species *versipellis* (or *floccopus* or *rufescens*), *scaber*, etc., the newer generic names for which are given above. It is believed that the use of the singular of this word for an entirely different group would accomplish no useful purpose and would only add confusion when it can easily be avoided.

Still another proposal involves a new genus. In the last few years, Murrill (**1938**, **1939** and **1940**) has described from Florida six new species in *Gyroporus* (the Cariosi of Fries and Peck), in four of which some important characters are at variance with the usual European conceptions of the genus. These species all have solid stipes. Further, the tubes of the species are not free as typically in *Gyroporus*. More important, however, the spores of these species are white in deposit instead of yellow, and instead of being broadly oblong-elliptical in shape are narrowly ellipsoid or cylindrical, if not more or less subfusiform—3 to 4 times longer than broad as compared with 1.5 to 2.5 times longer for the species of *Gyroporus* as most commonly conceived. Accordingly, it would seem that these would be better placed in a new genus.

Two other species of Murrill's make the situation not quite as clear-cut as one might wish it. *G. roseialbus* and *G. umbrinisqua-*

mosus produce spore-deposits that are white or nearly white instead of yellow, and the stipe of the former is solid. Since, however, the spores are broadly elliptical, they belong in *Gyroporus*.

The proposed new genus is as follows:

Leucogyroporus gen. nov.

Pileo sicco, e subtomentoso glabro; carne alba, non cyanescenti; tubulis e subdecurrentibus adnexis, non libris, albis vel pallidis, non flavescentibus, poris parvis; stipite glabro, solido; sporis anguste ellipsoideis, subfusiformibus vel cylindricis, in pulvere albis vel ochraceo-albis.

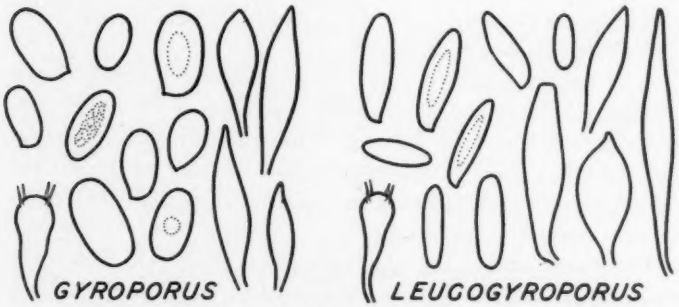

FIG. 1. Spores, cystidia and basidia of *Gyroporus* and *Leucogyroporus*. Spores × 1000; cystidia and basidia × 500.

Carpophores gymnocarpic. Surface dry, glabrous, felted to subtomentose. Flesh white, not changing to blue. Tubes adnexed, adnate, depressed or decurrent, not becoming free; white or pallid at first, remaining so or becoming isabelline to pale ochraceous or even reddish-brown in one species, not becoming yellow; pores small to minute. Stipe glabrous, even, solid. Spores narrowly ellipsoid to more or less cylindrical or subfusiform, 3 to 4 times longer than broad, white to ochraceous-white in deposit. Cystidia fusiform to fusiform-clavate, hyaline.

TYPE SPECIES—*Gyroporus pisciodorus* Murrill

The following new combinations are made: **Leucogyroporus pisciodorus** (Murr.) Snell comb. nov., **L. stramineus** (Murr.) Snell comb. nov., **L. Rhoadsiae** (Murr.) Snell comb. nov., and **L. deflexus** (Murr.) Snell comb. nov.

While one is on the subject of new genera of the Boletaceae, attention may be called to Murrill's new genus *Frostiella* (**1942**), erected to include two species with slender and coarsely reticulated or lacerated stipe and with ornamented spores—*Boletus Russellii* Frost and *B. Betula* Schw. In Gilbert's arrangement, these two species were placed in Murrill's genus *Boletellus* along with *B. Ananas* (*cf.* Snell, **1941**). In Murrill's scheme of the family, his new genus seems amply justified. Even in Gilbert's, there is much to be said for it, for either with or without two species added to *Boletellus* by the writer (*B. chrysenteroides* Snell and *B. subflavidus* Murrill—*loc. cit.*), the genus is more or less composite. Consideration has often been given to the splitting of *Boletellus* in the Gilbertian sense, but the temptation has been resisted in the interest of keeping down the number of genera with one or two species. Hence, for the time being at least, Murrill's *Frostiella* will not be added to a list of genera of the family already long and increased in this paper by one more new one.

SUMMARY

Since S. F. Gray's *Natural Arrangement of British Plants* has been found to be post-Friesian, it becomes desirable, if not necessary, to consider his generic names.

It is proposed to adopt *Suillus* Micheli ex S. F. Gray in place of *Ixocomus* Quélet, and *Leccinum* S. F. Gray in place of *Trachypus* Bataille.

It is also proposed to consider *Versipellis* Quélet a *nomen ambiguum* in order that it may not supersede *Xerocomus* Quélet, since the word Versipelles has long been associated with the *versipellis-scaber* group of species.

A new genus *Leucogyroporus* is proposed for four newly described Florida species originally placed in *Gyroporus* or the Friesian and Peckian Cariosi of *Boletus,* with tubes not free, stipe solid and spores narrowly elliptical and white or nearly white in deposit.

BROWN UNIVERSITY,
PROVIDENCE, R. I.

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PHOTOGRAPHS AND DESCRIPTIONS OF CUP-FUNGI—XXXVII. PEZICULA PURPURASCENS

FRED J. SEAVER

(WITH 1 FIGURE)

The above named species was originally collected at Westchester, Pennsylvania, in July, 1888, and distributed in North American Fungi *2147*. In working over the type material in The New York Botanical Garden, the writer was interested in noting a peculiar conidial stage associated with it, and possibly representing its conidial stage. The conidiophores resembled small asci and like the asci ruptured at the end permitting the four-celled conidium to emerge. This might be called an *Endoconidium* but, because of the close resemblance of the conidiophore to an ascus, the term **ascoconidiophore** is proposed, the conidium would then be an **ascoconidium.** Up to recently this species of *Pezicula* was apparently known only from the type collection in Pennsylvania.

In 1933, Dr. Theodore T. Ayers sent the writer a specimen collected by J. R. Hansbrough as *Dermatea purpurascens,* under which name it was originally described. Examination of this material on *Castanea dentata* showed the same ascus-like conidia found in the original collection made by Ellis, and since the two are constantly associated have good reason to assume that the two are organically connected.

This peculiar type of conidiophore is in itself sufficiently important to deserve special mention. Here we have a conidium which so closely simulates an ascus that the writer was at first led to wonder if there were not two ascomycetes associated. Careful examination, however, convinced us that the interesting structures were really conidiophores with the conidia produced internally. The ascoconidiophores are themselves pale brown, while the ascoconidia when released are nearly hyaline, or faintly smoky, and 3-septate, as are also the ascospores, but slightly different in form.

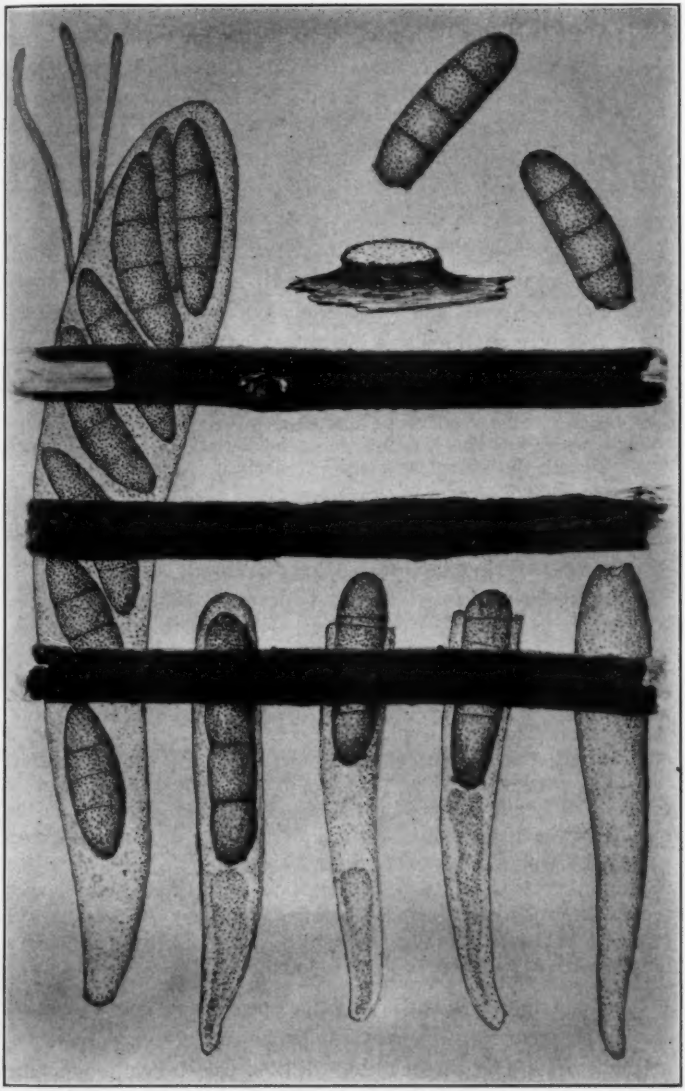

FIG. 1. *Pezicula purpurascens.*

We cannot, of course, claim that this is the conidial stage of the discomycete. However, it is remarkable that the two should be so closely associated in specimens collected more than forty years apart, the one in New Jersey and the other in Massachusetts. The conidia are produced in sori similar to those producing the apothecia and may precede them. Unfortunately the material is too old for cultural study, and the writer is presenting the facts as they are without attempting to explain them.

This fungus might be referred to the genus *Endoconidium* Prill. & Delac., but differs in that there is only a single 3-septate spore in each ascoconidium, and in that the ascoconidium is more ascus-like. It is here regarded as a distinct genus:

Ascoconidium Castaneae gen. et sp. nov.

Conidiophoris clavatis, ascorum similibus, fuligineis, 9–10 × 30–40 μ, monosporis; conidiis ellipsoideis, 3-septatis, subhyalinis.

On branches of *Castanea dentata* (Marsh.) Borkh. associated with *Pezicula purpurascens* (Ellis & Ev.) Seaver.

TYPE LOCALITY: Westchester, Pennsylvania.

DISTRIBUTION: Pennsylvania and Massachusetts.

Pezicula purpurascens (Ellis & Ev.) comb. nov.
Dermatea purpurascens Ellis & Ev. Jour. Myc. **4**: 100. 1888.

Apothecia scattered, erumpent, occurring singly or 2 or 3 crowded together, sessile or subsessile, externally reddish-purple, reaching a diameter of .75–1 mm.; hymenium plane or slightly concave, dirty-white becoming reddish-purple but lighter than the outside of the apothecium; asci cylindric-clavate, reaching a length of 120–140 μ and a diameter of 25–30 μ, 8-spored but some often undeveloped, 8–11 × 30–36 μ, hyaline or nearly so, ellipsoid with the ends strongly narrowed, becoming distinctly 3-septate, 9–11 × 30–36 μ; paraphyses slender, slightly enlarged above, reaching a diameter of 2–3 μ, often slightly colored.

Conidia found associated with this species and possibly representing its perfect stage. Ascoconidiophores club-shaped reaching a length of 90 μ and a diameter of 12 μ, pale brown, containing ascoconidia; ascoconidia broad ellipsoid reaching a length of 30–40 μ and a diameter of 9–10 μ borne on slender stalk within the

conidiophore becoming disconnected, and finally discharged through the ruptured conidiophore, 3-septate, appearing brownish within the conidiophore but hyaline or subhyaline when discharged.

The exterior of the apothecium is clothed with a palisade of appressed, poorly developed hairs which are dilutely purplish. It is this character which has suggested the specific name.

On dead limbs of chestnut, *Castanea dentata* (Marsh.) Borkh.

TYPE LOCALITY: West Chester, Pennsylvania.

DISTRIBUTION: Pennsylvania and Massachusetts.

EXSICCATI: N. Am. Fungi *2147*.

THE NEW YORK BOTANICAL GARDEN

EXPLANATION OF FIGURE

FIG. 1. *Pezicula purpurascens.* Photograph of chestnut branches bearing apothecia and conidia, about natural size. At the left, drawing of an ascus with spores and paraphyses. Above, drawing of one apothecium enlarged; also two ascoconidia. Below, *Ascoconidium Castaneae.* Several ascoconidiophores showing stages in the development and discharge of the ascoconidium. Photographed and drawn from type material collected at Westchester, Pennsylvania, in 1888.

STUDIES IN THE GENUS TRICHOLOMA—I

WALKER R. ARDE, JR.

(WITH 21 FIGURES)

During the past fifteen years the writer has been collecting fungi, mostly the hymenomycetes, in Pennsylvania, New Jersey and the New England States. Seven hundred and fifty paintings have been made from fresh material, spore observations made and data taken.

During the first years, the author was fortunate in having the help of Prof. Charles Kauffman and of Prof. Henry Beardslee. Also many of the *Russula* paintings were sent to Dr. Rene Maire in Morocco for his opinion.

During all this time very few new species were found. However, many so called "new species" were found to be identical with Friesian species.

In order to come to a conclusion on many controversal species it has been necessary to look up illustrations of nearly all the early mycologists that Fries referred to. In doubtful cases Fries' opinion, as stated in his Hymenomycetes of Europe, has been adhered to.

The *Tricholomae* have been dealt with first as they constitute a particularly difficult genus. So far, twenty-eight different species of *Tricholoma* have been found. Of these, twelve have not been reported from America before as given below but some of them have been reported under other names by American mycologists. In such cases the American synonym is given. Exsiccati of many are preserved in the New York Botanical Garden.

TRICHOLOMA QUINQUEPARTITUM Fries.

Agrees well with original figure *pl. 25* in Hymenomycetes of Europe.

Pileus 6 cm. broad, convex, umbonate (in some specimens), glabrous, *very viscid when wet,* lemon-yellow color with some

orange tints; lamellae adnexed, white, crowded, all lengths, 6 mm. broad; stipe tapering downward, ventricose in center, pallid with a faint yellow tint, crooked, *faintly discoloring, ferruginous where handled;* odor and taste farinaceous.

Solitary in birch and conifer woods at Moosehead Lake, Maine. Found on several occasions. Differs from *T. sejunctum* Fries in being viscid and in not having the pileus streaked with dark, fibrillose fibers. It nearly always has a crooked ventricose stem as Fries' illustration shows. New to America.

TRICHOLOMA ALBELLUM Fries (FIG. 3).

Pileus 5 to 8 cm. broad, hemispherical, then flattened, edge at first incurved, *striated with faint, fibrillose scales, often marked also with drop-like scales,* dry, pale fleshy-ochre color; lamellae emarginate, creamy-white to pale watery yellow, rather distant, brittle, thick at base; stipe short (5 cm.), thick (2 cm.), *usually with a thick, ovate bulb* that gradually merges into the stipe midway, *very firm,* chalky-white, *sordid-clay on handling;* odor strong, varying from nitrous, earthy, new-mown hay, etc.; taste mild; spores *minute,* $4\frac{1}{2}\ \mu \times 3\frac{1}{2}\ \mu$, white, nucleate.

Never in clusters as far as I have seen but usually gregarious. In coniferous woods at Valley Green, Penna. Also at Penn Valley 1934. Fries says in Hymenomycetes of Europe that there are two forms, one solitary as he found it and one caespitose which Sowerby described and which has not been found since. They seem like two different species to me. *T. albellum* and *T. gambosum* have often been confused. Fries says that *T. gambosum* always has an equal stem while *T. albellum* often has the base swollen by an ovate bulb. This is in contradiction to the name as *gambosum* means a swelled hoof. From Fries' earlier works it appears that what he first described as *gambosum* he later described as *T. albellum.* Apparently he later thought it was a form of *T. albellum* Sowerby. *T. gambosum* has more flesh-pink tones and always has an equal stem.

TRICHOLOMA LORICATUM Fries (FIG. 5).

Pileus large (9 cm.), convex-campanulate, then flattened, *with a thick, leather-like cuticle resembling parchment,* unpolished, greasy

when wet, livid-fuscous in center, pale yellow-green on edge, fading to dirty white; lamellae 7 cm. broad, adnate-emarginate, hardly crowded, very pale ochre; stipe 10 cm. long, 2½ cm. thick, equal, *hollow, fibrous, pale salmon color;* odor *spicy, disagreeable, like wild carrot;* taste somewhat farinaceous; spores white, smooth, 5–6 $\mu \times 3\frac{1}{2}\ \mu$, with an oil globule.

Found on several occasions growing in pairs, at Penn Valley, Pennsylvania, in frondose woods in October. This rare species has not been reported from America before. It has not been pictured by anyone as far as I know. The strong spicy odor makes it easy to identify. Hard's *pl. 63* as *T. saponaceum* looks like it. *T. saponaceum* Fries—its nearest neighbor is much smaller, has a short, tapering stipe, pileus polished and a soapy-farinaceous odor. Also the lamellae of *T. saponaceum* usually have a glaucous tinge.

TRICHOLOMA GUTTATUM Schaeffer (Stature of FIG. 8).

Pileus 6 cm. across, convex, edge somewhat incurved, *dry, somewhat squamose-scaly,* squamules often arranged in spot-like scales. Nearly cinnamon color (Ridgway), to vinaceous-drab, very firm; lamellae emarginate, 8 cm. broad, easily separating, close, dirty white color; stipe *solid, very firm,* naked (or a few fibrillose scales), bulbous, short (4 cm.), 2 cm. thick at base; odor none; taste at first little bitter and peppery, then mild; spores white, with an apiculus, 7 $\mu \times 5\frac{1}{2}\ \mu$, smooth.

Found growing solitary, at Penn Valley, Oct. 1933. Difficult to identify. *T. guttatum* is not well understood and is vaguely described. Fries says the specimens of Lasch showed the pileus to be more flocculose than granulose. Fries says it is cinnamon color.

TRICHOLOMA COLUMBETTA Fries f. *robusta* Sterbeeck.

Pileus *at first globular-convex,* edge incurved, *silky,* whitish, often with brick-red and grey spots; flesh thick; lamellae emarginate, 6 cm. broad, dirty cream color, brittle, often separating easily; stipe entirely bulbous, at first concolorous, very firm, solid; flesh or stipe inside slightly pink on cutting, in my specimens; odor none.

Grew in pairs at Penn Valley and Valley Green (coniferous woods), fall 1925.

TRICHOLOMA SPERMATICUM Paulet (FIG. 10).

Pileus 9 cm. broad, convex-flattened, with a rounded umbus, edge incurved, chalky white, umbus little buff, not viscid, apparently hygrophanous; flesh snow-white, unchanging; lamellae notched and free, 8 mm. broad, close, thin, *somewhat ragged on the edge,* pale dirty cream-straw color; stipe long (9 cm.), thin (1 cm.), equal, solid, distinctly cartilaginous, little furfaceous near top, white mycelium at base; odor farinaceous-disagreeable and semen-like at the same time; taste first farinaceous then peppery; spores round to ovate-round, hyaline, $6\frac{1}{2}\ \mu \times 5\frac{1}{2}\ \mu$.

Growing in pairs, on ground, in frondose woods, Penn Valley. Not reported from America before to my knowledge.

TRICHOLOMA INAMOENUM Fries (FIG. 12).

Pileus 5 cm. broad, thick, hemispherical-umbonate, silky-smooth, dry, dingy-white; flesh thick, pure-white; lamellae very broad toward stipe, *with a decurrent tooth distant, thick* shiny-white with a cream-flesh tint; stipe long (7 cm.), 12 mm. thick, equal, but with a slight knob that is buried, pure white, solid; odor not pronounced; taste strongly farinaceous; spores smooth, $4\frac{1}{2}\ \mu \times 3\frac{1}{2}\ \mu$.

Deeply *imbedded in pine needles,* Bryn Mawr. New to America.

TRICHOLOMA BREVIPES Bulliard.

Pileus 8 cm. broad, flat, with undulating-drooping edge, soft, chamois-like feeling, dry, creamy-white; lamellae *crowded,* creamy-yellow, emarginate-free, some forked and many shorter ones, 7 mm. broad, *they stop short of the stipe;* stipe short ($2\frac{1}{2}$ cm.), knob-like on end, *tough due to outer fibrous coat,* concolorous, but *internally ropy and ferruginous tinted,* strongly attached; odor none; taste bitter in my specimens.

Found growing solitary on ground, in open woods, near Toronto, Canada. New to America. The outstanding feature (as Bulliard shows in his plate) is the short stipe that is ferruginous inside.

TRICHOLOMA PORTENTOSUM Fries.

Pileus 7 cm. across, convex, firm, not viscid, ecru-drab (Ridg.), *streaked with black, fibrillose lines,* sometimes arranged in the form of scales; lamellae rounded, almost free, whitish at first, *then pale drab;* stipe *short, very firm,* solid, smooth, white inside and outside,

somewhat bulbous, deeply imbedded in soil, odor none; taste peppery (in my specimens).

Grew solitary, on the ground, in frondose woods. Fries' *Pl. 24* in Hymenomycetes shows colors well. It is a fuliginous-livid color.

TRICHOLOMA PUTIDUM Fries (FIG. 15).

Pileus 4 cm. broad, hemispherical, thin, flaccid, *soft, chamois-like feeling,* pale greyish-olive (deep olive-buff) Ridg., hygrophanous, somewhat hoary; lamellae also olive-buff, sinuate-adnate, *distant,* broad (6 mm.); stipe 4 cm. long, 6 mm. thick, equal, fibrous outer coat, *often flattened;* odor very disagreeable (when old); spores pip-shaped, $7\ \mu \times 3\frac{1}{2}\ \mu$.

Found growing solitary on buried wood-chips, at base of rotten stump, at Valley Green, in coniferous woods. The cap is somewhat velvety. Resembles Cooke's *Pl. 601* of *Naucoria centunculus.* New to America.

TRICHOLOMA LURIDUM Schaeffer (Stature of FIG. 17).

Synonym—*Tricholoma duracinum* Cooke.

Pileus $4\frac{1}{2}$ cm. broad, convex, *irregular and lobed,* edge incurved, silky, somewhat fibrillose, serpentine-green (Webster's Dict.), and pale testaceous, becoming streaked with cinereous; lamellae emarginate, crowded, pale olive-grey (griseus—Sacc.), edge white-fimbriate, all lengths; stipe short (4 cm.), thicker and curved at base (2 cm.), solid, cartilaginous, fibrillose, with rusty-brown stains; odor earthy pleasant; taste mild (but a little warm after-taste); spores, $6\frac{1}{2}\ \mu \times 5\ \mu$, greenish-brown under the microscope.

Grew solitary in frondose woods, Penn Valley, rare. New to America. Cooke's *Pl. 214* Ill. of British Fungi shows it well.

Tricholoma cuneifoloides Arde, sp. nov. (FIG. 20).

Pileus unciam latus, convexo-obtuso, isabellinus (novus solanum tuberosum color), laevi-punctato vel virgato cum minima-squamae, *glutinosis;* lamellae cuneus-formus, albis; stipe albis, levis, cuticula cartilaginea, $2\frac{1}{2}$ cm. longus, sporus levis, globus, albis, $5\frac{1}{2}\ \mu$.

With *T. cuneifolium* Fries it is close but differs in the very viscid pileus.

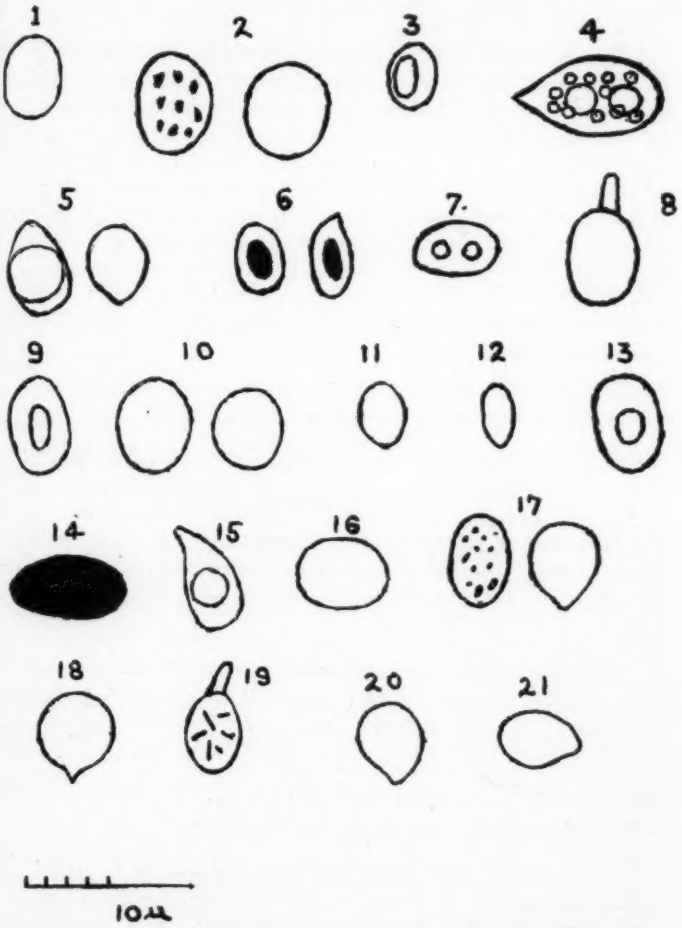

FIG. 1, *Tricholoma flavobrunneum;* 2, *T. equestre;* 3, *T. albellum;* 4, *T. sulphureum;* 5, *T. loricatum;* 6, *T. saponaceum;* 7, *T. amethystinum;* 8, *T. guttatum;* 9, *T. panaeolum;* 10, *T. spermaticum;* 11, *T. album;* 12, *T. inamoenum;* 13, *T. argyraceum;* 14, *T. virgatum;* 15, *T. putidum;* 16, *T. rutilans;* 17, *T. luridum;* 18, *T. cinerascens;* 19, *T. acerbum;* 20, *T. cuneifoloides;* 21, *T. subsejunctum.*

TRICHOLOMA VIRGATUM Fries (FIG. 14).

Pileus 5 cm. broad, convex-hemispherical, thick, smoke-grey (Ridg.), *densely virgate with small, fibrillose scales,* dry; flesh thick, white, but *soon moddled grey;* lamellae sinuate, not as thick as the pileus (5 mm.), close, *smoke-grey,* darker on the edge, shorter ones; stipe equal but twisted, 5 cm. long, 5 mm. broad, silky-fibrillose (bark-like), also pale, smoke-grey, solid, little pink at base; odor strongly farinaceous; taste *very bitter;* spores elliptical, smooth, $8 \times 5\ \mu$, purple-brown under the microscope.

Found growing solitary on a bank in coniferous woods. Seems close to *T. murinaceum* Bulliard but that species is not definitely said to have a bitter taste.

TRICHOLOMA AMETHYSTINUM Scopoli (FIG. 7).

Pileus 6 cm. broad, convex-plane, mottled, color *lateritius* (Sacc.), with azure-blue spots, edge pale glaucous-cream, hardly viscid; lamellate rounded-adnexed, ventricose distinct, rather distant, soft, watery, cream-flesh color; stipe *very fragile, soon hollow,* fibrous (breaking in shreads), pure-white, but sordid on handling; odor and taste mild; spores white, smooth, oval, $5\frac{1}{2}\ \mu \times 3\ \mu$, two guttate.

Found growing solitary at Penn Valley. Rare. Not well known. Fries seems to have confused it with *T. lixivius* which is ash color. This is amethyst color.

TRICHOLOMA SULPHUREUM Bulliard (FIG. 4).

Synonym—*Tricholoma chrysenteroides* Peck.

Pileus 4 cm. broad, campanulate, pale buff-yellow, unpolished; lamellae adnate, pallid, 6 mm. broad, close; stipe equal, *tortuous,* roapy, sulphur yellow (inside and outside); odor farinaceous; taste almond; spores almond-shape, sculptured, $9 \times 5\ \mu$.

After examining Bulliard's *Pl. 168* of the above I see no reason why *T. chrysenteroides* Peck is not the same.

Just a word concerning some other difficult species that have been found here.

TRICHOLOMA ACERBUM Bulliard (FIG. 19).

Exactly resembles Bulliard's *Pl. 571* of same. It grows in clusters, has large thick caps, buckthorn-brown (Ridg.) with center vinaceous-red. Edge of cap incurved. Lamellae *very narrow*, sulphur-yellow mycelium at base of stipe; odor strong, fungus-like, taste bitter.

TRICHOLOMA CINERASCENS Bulliard (FIG. 18).

Illustrations misleading. Boudier's *Pl. 29* (Icones Mycologicae) shows it best. Pileus is pale ochre, dry, with a soft feeling, very tough at first but quite brittle on drying, edge turned up somewhat and undulating; lamellae attenuated at both ends, crowded, cream-straw color; odor strong (horse-radish); taste mild to slightly bitter; spores globose, white, smooth, 5 μ (again 5 $\times$ 3 μ).

Grows in clusters on the ground in the spring.

TRICHOLOMA ALBUM Fries (FIG. 11).

My specimens resembled Kauffman's *Pl. 151* (of *T. acerbum*). Pileus convex-gibbous, thick in center, ivory-soap color, somewhat hoary and mottled, not viscid; lamellae very crowded, dirty-white; stipe with an ovate-bulbous base, short; odor none; taste farinaceous at first, then sharp or sharp-bitter; spores minute, 5 $\times$ 3 μ, smooth.

TRICHOLOMA SAPONACEUM Fries (FIG. 6).

Stature of figure 6 but with a thinner stem. Pileus polished, pale fuscous, colors often stippled edge pale olive, semiviscid; lamellae, ventricose, cream color with a glaucous tinge, stipe short, white but with a flesh tint inside; taste farinaceous; odor soapy as in a laundry; spores white with a pink nucleus, 4 $\times$ 3 μ.

Common here after frosts.

The author is indebted to Dr. Seaver for his encouraging hand and for placing the very complete library of the New York Botanical Garden at his disposal.

302 NORTH 40TH STREET,
PHILADELPHIA, PA.

CHROMOBLASTOMYCOSIS [1]

A. L. CARRIÓN

(WITH 7 FIGURES)

Definition. Chromoblastomycosis is a chronic, infectious, apparently non-contagious, granulomatous dermatosis which is usually confined to an exposed area of the skin and may be caused by different but closely related dematiaceous fungi.

Geographic distribution. The disease was discovered by A. Pedroso, of Brazil, in 1911 (**1**), but the first case appearing in the literature occurred in Boston and was published by Medlar and Lane in 1915 (**2, 3**). Since that time at least one hundred authentic cases of chromoblastomycosis have been recognized throughout the world. The records would point to Brazil and Puerto Rico as the most heavily infected foci in existence, but the true incidence of this disease will not be known until the medical profession in different countries becomes better acquainted with the pathologic process and with the methods of diagnosis.

Etiologic factors. Chromoblastomycosis is most common during the period of active adult life; the higher incidence falls upon males and there seems to be no race immunity. As a rule, the patients give a history of being farm laborers who work barefooted most of the time. Transmission from man to man has never been recorded so far. Apparently, the fungus is present in the soil and individuals who are susceptible contract the infection through some unimportant abrasion of the skin.

Clinical aspects. The disease usually starts as a small, warty growth in some part of the foot, whence it extends upward through the development of satellite lesions. The course of the pathologic process is extremely slow, the duration at the time of examination often having been ten or twelve years.

An advanced case of chromoblastomycosis offers a most extraordinary dermatological picture (FIG. 1). The lesions occur in

[1] This study was made possible by a grant from the Bailey K. Ashford Fund.

great numbers and there is usually a certain degree of elephantiasis of the affected limb. The morphology of the lesions is extremely varied. Some of them consist of hard, elevated, variously sized, dull-pink or violaceous nodules, the surface of which is often irregular, verrucous and scaly. Larger lesions take the form of markedly prominent, sessile or pedunculated, cauliflower-like tumors. Superficially, these tumors are covered with a thick layer of hyperkeratotic epithelium which often falls off, exposing large numbers of pink papillomata. In a third type of lesion, the pathologic process forms moderately elevated, dull-red, scaly patches or plaques. Some of these plaques have a flat surface and may show exaggeration of the lines of cleavage of the skin; others become irregular due to the development, within the plaque, of papillomatous or nodular efflorescences, and still others, tend to heal centrally with the production of profuse scarring, the borders remaining active. Finally, there is a fourth type of lesion consisting of discrete or diffuse, hyperkeratotic growths that are purely verrucous in character.

The lesions of chromoblastomycosis are easily traumatized, they bleed readily, they may be complicated with bacterial infection and ulceration, and their surface often shows crusting and epidermal débris. Subjectively, pruritus is frequently an important symptom. Some patients complain of pain and, in advanced cases, there is partial incapacity for work. The deeper tissues are not usually involved, although there is one instance in which the bones of the leg were presumably affected. The lymphatic glands, draining the diseased focus, may participate in the process, but this is not the rule. However, adenitis due to bacterial complications, occurs frequently in patients with chromoblastomycosis. Metastases through the blood stream appear to be extremely rare, but there is no question that they can be produced (**4**). Finally, no systemic symptoms have yet been recorded for this disease.

It should be emphasized that the clinical picture just given is that of a well-developed, typical case of chromoblastomycosis. This picture is subject to variation. An early infection, one which has lasted for two or three years, for example, may consist of a single or a few verrucous growths, nodules or patches, which may not be specifically characteristic of chromoblastomycosis. On the

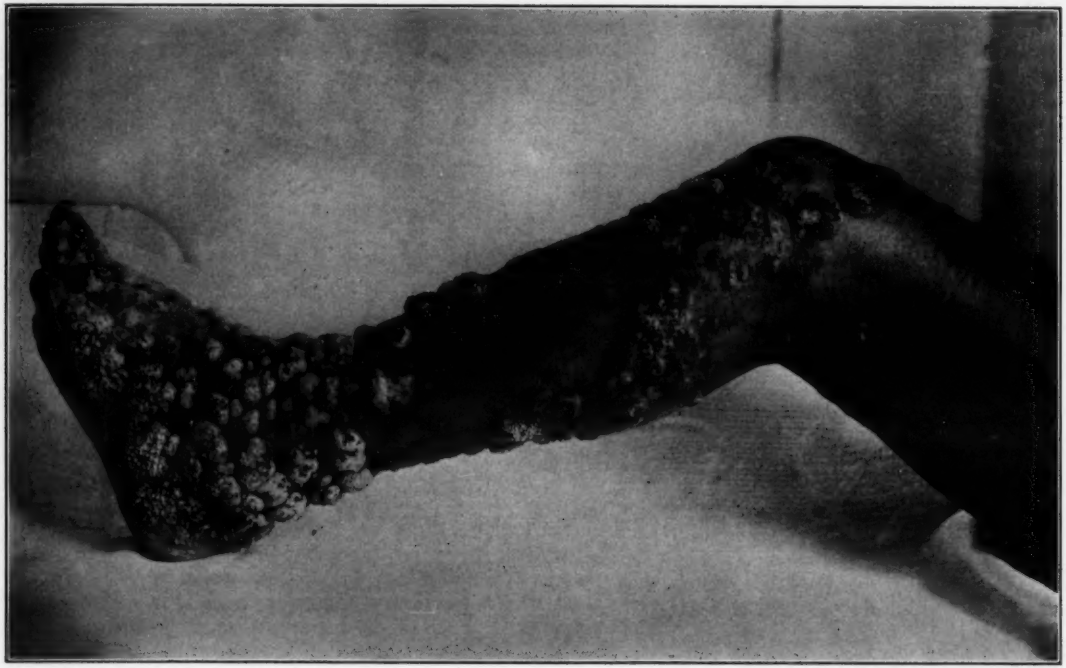

FIG. 1. Chromoblastomycosis in a male, white, Puerto Rican farm laborer, who contracted the infection fifteen years prior to examination while working barefooted in a coffee plantation.

other hand, in certain late infections, the lesions have become stationary before extending to any considerable degree. In such instances there may be found one or more ulcerated nodules, a papillomatous patch or a pseudo-ulcer with a depressed central area of hard, fibrotic scar tissue surrounded by an elevated, granulomatous growth.

Location may also influence the clinical picture. The large cauliflower-like tumors already described occur characteristically on the foot and lower leg. As a rule, the higher a lesion is located on the extremity, the less it will be elevated above the surrounding skin. Consequently, on the upper leg and thigh, nodules and plaques tend to predominate. Lesions of purely verrucous type are most frequently encountered on the foot, especially toward the borders and on the sole. Chromoblastomycosis is not nearly as frequent on the upper as on the lower extremities and the tumor lesions at the former location have never been as large as those often noted on the legs and feet. Otherwise the eruption is similar in both regions. Infections of the face have always been small and of the plaque type.

The species of fungus causing the infection does not seem to have much influence on the clinical picture. A possible exception was a Puerto Rican case in which the disease was confined to an upper limb and the lesions consisted of extensive, diffuse, even areas of infiltration with some papillomata on the hand and without tumors or nodules (**5**). This was the case produced by *Fonsecaea compactum.*

Histopathology. Histopathologically, chromoblastomycosis is a typical infectious granuloma. The lesions affect the epidermis, the cutis and the subcutaneous tissues and they tend to develop toward the surface with very little disturbance of the deeper structures. Microscopic examination reveals a dense cellular infiltration which includes lymphocytes, plasma cells, polymorphonuclears, eosinophiles, epithelioid cells and occasional giant cells of the Langhan's type. The pathologic reaction may be focal or diffuse; in places, it is distinctly tuberculoid; there is marked capillary engorgement as well as edema of the tissues and, not infrequently, microscopic abscesses. A constant feature of chromoblastomycosis is the development of an intense fibrosis which tends to wall off the infected

foci. The parasites are observed either within the giant cells or free in the tissues as spherical bodies measuring about 12 microns in diameter. These bodies, or sclerotic cells as they are often called, possess a dark, thick membrane and they often show internal septation. Their protoplasm is olivaceous and granular. In the epidermis there is usually a pronounced hyperkeratosis and acanthosis with the hypertrophic rete layer often forming irregular and interlacing epithelial columns which may extend deeply into the corium (FIG. 2).

Diagnosis. There are four essential elements of diagnosis in chromoblastomycosis. First among these is, of course, the clinical picture. This may be so characteristic that a mere look at the patient has often led to the correct diagnosis. However, there are other diseases, such as leprosy, syphilis, tuberculosis, mossy foot, etc., with which chromoblastomycosis might be confused and, consequently, laboratory investigations should always follow clinical inspection. It is worth mentioning that we have seen exceptional cases in which the eruption was as extensive and as typical as in any good case of chromoblastomycosis and, yet, it was impossible to demonstrate the presence of an etiologic agent of any sort. Whether these represent cured cases of the disease or some other infection of obscure etiology, it has been impossible to determine so far.

The second diagnostic element is the presence of typical sclerotic cells in the epidermal débris obtained by scraping the lesions. This is ascertained through microscopic examination of the material after mounting in a 40 per cent solution of potassium hydroxide (FIG. 3). Diagnosis should be verified further with a biopsy showing the characteristic histopathologic changes and the presence of the parasite and, finally, with the identification of the causative fungus in cultures from the scrapings or infected tissues.

Mycology. We do not propose to discuss here in full what is known about the fungi of chromoblastomycosis. It is our only purpose to focus the subject broadly and to present a brief summary of its fundamental highlights.

Repeated observations on a large number of isolates from cases of chromoblastomycosis in different parts of the World indicate that sporulation in this pathogenic group may be of three distinct

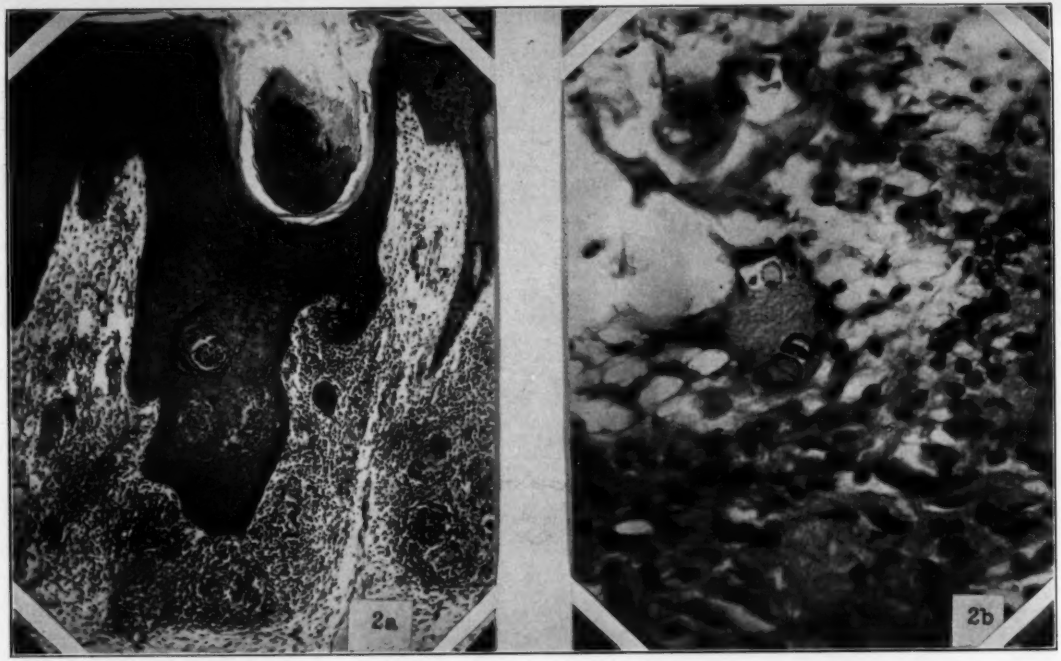

FIG. 2. Histopathologic sections of a lesion of chromoblastomycosis showing, at *"a,"* epidermal changes and granulomatous infiltration of the dermis, and at *"b,"* parasitic cells (so-called sclerotic cells) within a giant cell. Note internal septation in one of the fungus cells.

types. One of these types is the *Hormodendrum* (*Cladosporium*), in which the conidia are produced acrogenously in arborescent chains on the mycelial branches (FIG. 4: *a* and *b*). In the second, or *Phialophora* type, the sporulation is semi-endogenous in nature. The conidiophore is a flask-shaped cell and the spores bud out in succession from the constricted portion of the cell into an adjacent cup where they are often glued together forming characteristic spherical masses (FIG. 4: *c* and *d*). Finally, the *Acrotheca* method of sporulation is characterized by a specialized conidiophore consisting of a more or less extensive, straight or irregular, hyperpigmented, sometimes swollen hyphal segment which may be disposed terminally, intercalarily, or as a lateral branch. A conspicuous feature of this conidiophore is the presence throughout its surface of a large number of tiny, truncate, conical processes, to which the exogenous spores are united and which give to the fruiting structure a very characteristic verrucous appearance. Some of these conidiophores do not extend very much in length, but take the form of a short, swollen, irregular, warty growth. In certain strains in which this method of sporulation has reached its highest degree of development, the conidia are borne singly and only occasionally are secondary spores produced, forming chains of two (FIG. 4: *e* and *f*). In other specimens, however, spore heads of the *Acrotheca* type may show a greater tendency to chain formation.

A few of the fungi of chromoblastomycosis would seem to sporulate exclusively by one of the above methods. This is true of the species known as *Phialophora verrucosa* Medlar 1915 (**6**), in which the conidia are borne semi-endogenously in the form already described (FIG. 4: *c* and *d*). This is also true of a *Hormodendrum* species recently isolated by J. A. O'Daly [2] in a Venezuelan case. In this *Hormodendrum* we have been unable to find the *Phialophora* or the *Acrotheca*-like sporulations (FIG. 4: *b*). In most of the fungi of chromoblastomycosis, however, the three methods of sporulation, or at least two of them, occur simultaneously in the individual isolates (**7**). The organisms behaving in this manner

[2] Personal communication to the author. Doctor J. A. O'Daly kindly sent us a culture of this organism for study. As far as we know, the fungus has not been described as yet.

have been classed in two different species of the genus *Fonsecaea,* namely, *F. Pedrosoi* (Brumpt) Negroni, 1936, emend (**8**) and *F. compactum* Carrión, 1935, emend 1940 (**5, 7**). *Fonsecaea compactum* is represented by only one isolate discovered in Puerto Rico a few years ago. On the other hand, *Fonsecaea Pedrosoi* constitutes the vast majority of the fungi of chromoblastomycosis.

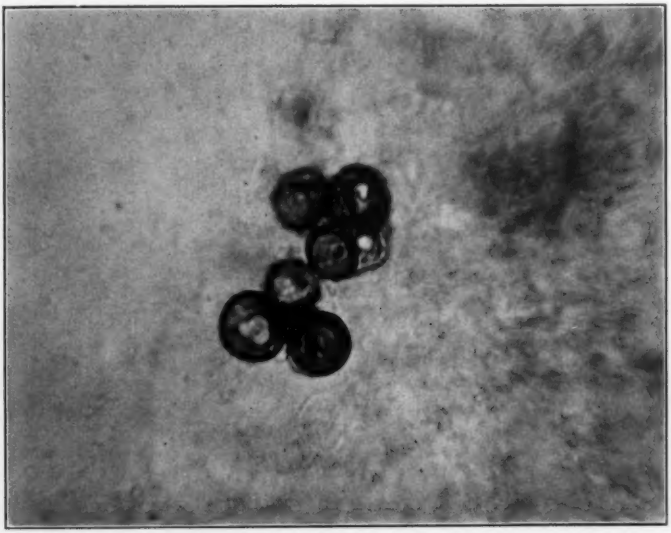

FIG. 3. Typical fungus cells found in the epidermal debris in a lesion of chromoblastomycosis.

The different types of sporulation characteristic of *Fonsecaea Pedrosoi* do not occur in the same proportions in all the strains of that species. In order to avoid confusion, therefore, it has been necessary to subdivide the group into a number of varieties in accordance with the predominant method of sporulation (**7**). *Fonsecaea Pedrosoi typicus* corresponds morphologically with Brumpt's original description of the fungus (**10**). Here the *Acrotheca*-like sporulation reaches its highest degree of development as to both quality and abundance. In members of this variety, the *Hormodendrum* heads may be scant, abnormal or depauperate.

The *Phialophora* cups are also very rare or missing. It would seem that both the *Hormodendrum* and the *Phialophora* methods of sporulation are becoming extinct in *F. Pedrosoi typicus* (FIG. 5: *a* to *f*).

In the second variety of *Pedrosoi*, namely, *Cladosporioides*, there is a similar situation, except that *Hormodendrum* (*Cladosporium*) is here the predominant character. In certain specimens of this variety, it is extremely hard to find sporulation of the *Acrotheca* and *Phialophora* types. In this instance it would seem that the *Acrotheca* and *Phialophora* methods are becoming extinct (FIG. 5: *g, h, i*).

In a third variety, *Phialophorica*, the *Phialophora* method is the preponderant. The only known isolate of this variety, originally described as *Phialophora macrospora*, produces typical spore heads of the *Acrotheca* type, while the *Hormodendrum* has become apparently extinct (FIG. 6).

Finally, the variety *F. Pedrosoi communis* reveals the three methods of sporulation in more or less conspicuous abundance. *Pedrosoi communis* includes a large number of intergrading forms which represent connecting links among the other three varieties (FIG. 5: *j, k, l*).

According to these observations it would seem that the fungi of chromoblastomycosis have all a common origin, namely, the variety *F. Pedrosoi communis*, which possesses the three methods of sporulation. Following different lines of evolution, certain strains of this group have gradually lost their ability to sporulate by any one or two of these methods. Thus, in the species *Phialophora verrucosa*, the *Phialophora* is the only method retained. It becomes evident, moreover, that the variety *F. Pedrosoi Phialophorica* represents a transition between the original group *F. Pedrosoi communis* and *Phialophora verrucosa*. Similarly, in the *Hormodendrum* species from Venezuela (O'Daly), only the *Hormodendrum* type has been retained, the variety *F. Pedrosoi Cladosporioides* representing the transitional group in this instance. Up to the present time, none of the fungi of chromoblastomycosis have been found to sporulate exclusively by the *Acrotheca* method, but we should not be surprised if, in the future, new isolates are discovered in which this method is the only one observed. The specimen de-

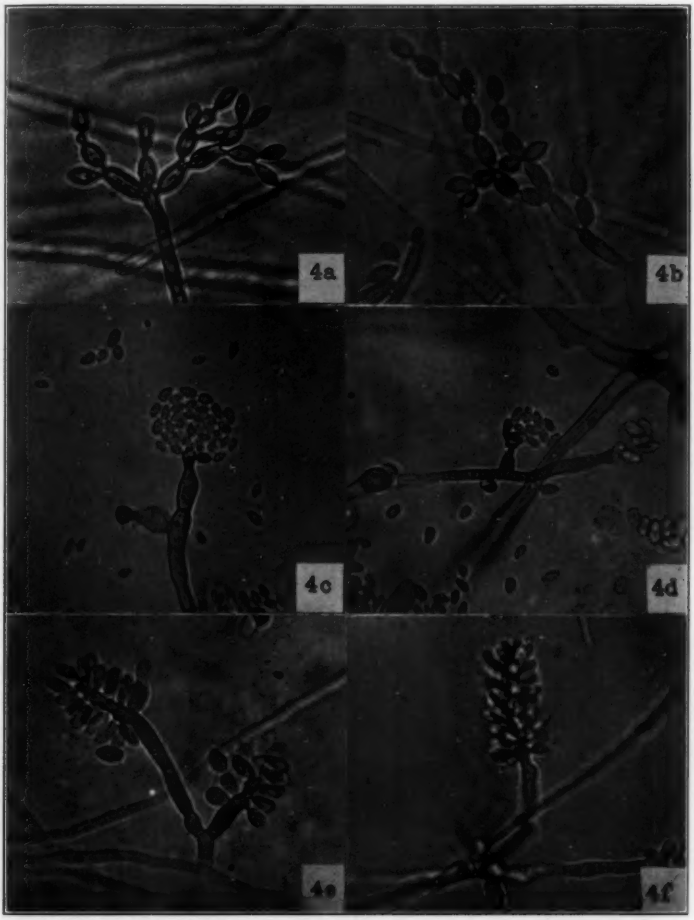

FIG. 4. Different methods of sporulation noted in fungi of chromoblastomycosis: *a*, *Hormodendrum* sporulation in a specimen of *Fonsecaea Pedrosoi communis* isolated from a case of chromoblastomycosis in the Dominican Republic; *b*, typical spore head in a *Hormodendrum* species isolated from a Venezuelan case; *c* and *d*, semi-endogenous sporulation in *Phialophora verrucosa* (Uruguayan isolate); *e*, *Acrotheca* type of sporulation in *Fonsecaea Pedrosoi communis* (Philadelphia case); *f*, *Acrotheca*-like sporulation in a Brazilian isolate of *Fonsecaea Pedrosoi typicus* (the so-called *Hormodendroides Pedrosoi*).

scribed as *Botrytoides monophora* (**15**) comes very close to fulfilling this condition (FIG. 7).

The classification of the fungus *Fonsecaea Pedrosoi* with its complicated morphology has been a much debated subject. The differences of opinion have centered essentially on two questions: 1. Are we dealing with only one species, namely, *Pedrosoi,* or with several? 2. Under what genus should *Pedrosoi* be placed?

Different varieties of *Fonsecaea Pedrosoi* have been repeatedly described as independent species by many investigators who have misinterpreted or over-emphasized the importance of one or another of the morphologic features of that fungus. Consequently, a review of the literature on the mycology of chromoblastomycosis will reveal a large number of specific names given to individual isolates which, apparently, did not correspond to the original description of *Pedrosoi* (**10**). However, after many years of careful and patient work, it has been demonstrated: (*a*) that the fungi described under such names have all a common tendency to sporulate by the three methods characteristic of *Pedrosoi* and cannot be considered as independent species; (*b*) that the only differences existing among these organisms lie in the comparative proportions in which these methods occur; and, finally, that these differences are quantitative rather than qualitative and, therefore, the only subdivision possible in this group should fall in the rank of varieties (**7**).

The second point of debate about the species *Pedrosoi* has been its proper generic name. As already stated, most of the names proposed for this species in the past have fallen into synonymy. However, there is still difference of opinion as to whether the fungus in question should be classed among the Hormodendrums, the Phialophoras or the Fonsecaeas.

Shall *Hormodendrum* be retained? *Hormodendrum* is supported by the rule of priority and by the fact that a large number of isolates of the varieties *Cladosporioides* and *communis* present the *Hormodendrum* sporulation as an outstanding or, at least, a conspicuous character. On the other hand, there are fundamental objections to the use of that name. In the first place, the genus *Hormodendrum* would not admit certain isolates of the varieties *typicus* and *Phialophorica* because, in these isolates, the *Hormo-*

dendrum sporulation has become more or less obsolete while other methods of reproduction, typical of other well established genera, predominate. In the second place, experience has shown that the application of the name *Hormodendrum* to the species *Pedrosoi* is responsible for most of the confusion that has hitherto existed among the fungi of chromoblastomycosis. It is the objection to this name that has moved such a large group of investigators to place this fungus in so many other genera, leading to the long list of synonyms found in the literature. When Ota (**11**) and Langeron (**12**) erroneously placed *Pedrosoi* among the Trichosporiums, they were evidently impressed by the *Acrotheca*-like clusters, which were mistaken for *Trichosporium*, and they paid little or no attention to the scant and depauperate *Hormodendrum* sporulation noted in their cultures. Similarly, the generic names *Acrotheca* (**13**), *Gomphinaria* (**14**), *Botrytoides* (**15**) and *Hormodendroides* (**15**) have been applied to the species *Pedrosoi* by other well-known investigators who also placed the emphasis on the spore clusters of *Acrotheca* type, although they recognized the presence of *Hormodendrum* sporulation. In all these instances, it is clear that the authors were dealing with specimens of *Fonsecaea Pedrosoi typicus*. Even Brumpt, who is the author of the species *Pedrosoi*, and who called it a *Hormodendrum* in his original description in 1922 (**10**), was forced to admit later that the *Hormodendrum* sporulation was not the important character in the fungus he had studied (**16**).

Other workers have emphasized the importance of the semi-endogenous sporulation observed in *Pedrosoi* and have proposed two additional generic denominations for this species, namely, *Phialoconidiophora* (**15**) and *Phialophora* (**17**). These authors, too, have considered the *Hormodendrum* sporulation a feature of secondary importance. Finally, it is evident that Negroni, of Buenos Aires, had the same point of view when he called the fungus a *Fonsecaea* (**8**).

Summing up the situation, here are eight different generic names applied to one and the same species by a dozen different investigators who think that this fungus should not fall among the Hormodendrums. Indeed, this *is* confusion. On the basis of this experience and in compliance with one of the most essential

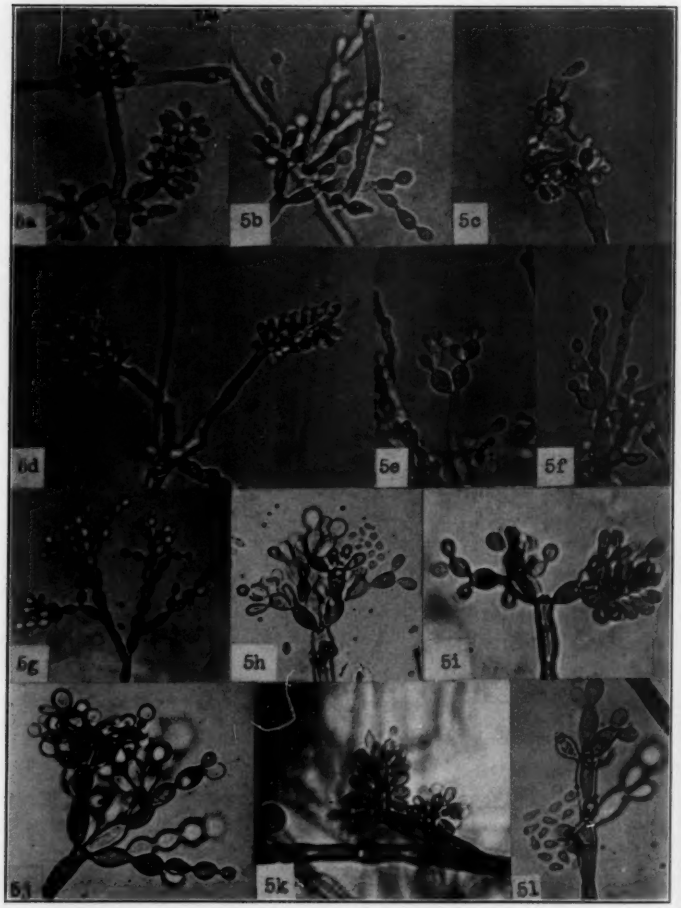

FIG. 5. *a*, *b* and *c*, *Acrotheca*-like, *Hormodendum* and *Phialophora* types of sporulation in a South American isolate of *Fonsecaea Pedrosoi typicus* (the so-called *Botrytoides monophora*); *d*, *e* and *f*, the three types of sporulation in another South American isolate of the variety *typicus* (the so-called *Hormodendroides Pedrosoi*); *g*, *h* and *i*, triple sporulation in a South American strain, *Fonsecaea Pedrosoi Cladosporioides* (the so-called *Phialoconidiophora Guggenheimia*); *j*, *k* and *l*, sporulation of the three types in a Puerto Rican isolate of the variety *F. Pedrosoi communis*.

principles of nomenclature, which is "to avoid or reject the use of . . . names which may cause error or ambiguity or throw science into confusion" (**18**), the generic name *Hormodendrum* should be eliminated in this case.

Shall it be *Phialophora?* In the present state of our knowledge, the inclusion of *Pedrosoi* in the genus *Phialophora* would be objectionable for reasons fundamentally similar to those given against

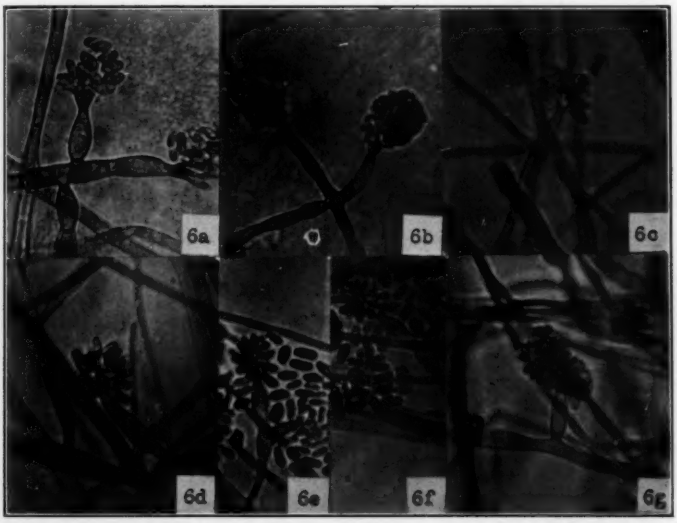

FIG. 6. The only known isolate of *Fonsecaea Pedrosoi Phialophorica* (the so-called *Phialophora macrospora*) obtained from a South American case. Note the typical sporulation of *Phialophora* in *"a,"* and *"b,"* and the *Acrotheca*-like sporulation in *"c," "d," "e," "f,"* and *"g."*

Hormodendrum. Indeed, *Phialophora* would be more confusing than *Hormodendrum.* Among the numerous specimens of *Pedrosoi* so far isolated and studied, there is only one in which the *Phialophora* sporulation predominates; in all the rest, there is an overwhelming preponderance of either the *Hormodendrum* or the *Acrotheca* methods of reproduction. Under such circumstances, *Phialophora* would be a poor substitute for *Hormodendrum.* A change in nomenclature is not justified unless the new name has substantial advantage over the old.

Why *Fonsecaea?* For several years we used consistently the binomial *Hormodendrum Pedrosoi.* At the present time we are calling this fungus a *Fonsecaea,* not because we like *Fonsecaea* better, but because we feel that, using this name, the busy students in medical mycology can work more effectively, lose less time and understand each other better. Notwithstanding this, if the men working in this field should get together, discuss and come to an

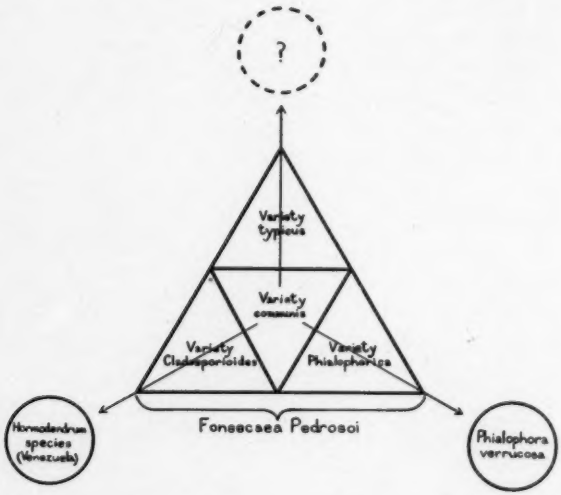

FIG. 7. Diagram showing apparent interrelations among certain fungi of chromoblastomycosis. The large triangle represents *Fonsecaea Pedrosoi,* which covers most of the fungi isolated from that disease. The included smaller triangles represent the four varieties which make up the species *Pedrosoi.* The arrows indicate different lines of evolution suggested. The circles represent independent species either real or potential. In *Fonsecaea Pedrosoi,* the variety *communis,* which possesses the three types of sporulation—*Cladosporium, Phialophora* and *Acrotheca,*—appears to be the common origin of all the other forms. The varieties *Cladosporioides, typicus* and *Phialophorica* show, respectively, a marked predominance of the *Cladosporium, Acrotheca* or *Phialophora* sporulations with a corresponding reduction, in each case, of the other two methods of reproduction. In the species *Phialophora verrucosa* and in the *Hormodendrum* isolate from Venezuela (see circles), the *Phialophora* and the *Cladosporium,* respectively, have become the exclusive methods of reproduction. The broken-line circle would represent the presumptive existence of other parasites sporulating exclusively by the *Acrotheca* method.

agreement on this point of nomenclature, we would be glad to abide by the decision of the group, no matter what that decision might be. Up to the present time, however, the more authoritative mycologists have failed to agree on this important subject. Under such circumstances, and with a purely compromising spirit, we have accepted the generic name *Fonsecaea* as a good substitute for *Hormodendrum* in the case of *Pedrosoi*. *Fonsecaea* is a legitimately created and comprehensive genus which covers, without strain, all the varieties of the species *Pedrosoi*. Its creation has solved a situation for which there is no adequate provision in the International Rules of Botanical Nomenclature. It represents a mycologic group possessing distinct pathogenic properties. As a name it is neither misleading nor confusing. We grant that *Fonsecaea* may not be a permanent generic name for the species *Pedrosoi,* but *Hormodendrum* and *Phialophora,* which are also imperfect genera, are not permanent either. Indeed, *Hormodendrum* is worse than *Fonsecaea* in this respect because, according to many well-known authorities, it should be replaced by *Cladosporium*. The correct botanical classification of *Pedrosoi* will be definitely established only when its perfect form becomes known, but there is no way of estimating how long a period will elapse before the sexual phase of this parasite is discovered, if discovered at all. In the meantime, it would seem unscientific and inconsistent with one of the fundamental principles of nomenclature to preserve a name which experience has proved to be a permanent source of error, ambiguity and confusion. With the new and broader conception of the species *Pedrosoi* including its different varieties, and with a generic name that is not confusing nor misleading, the classification of any fungus isolated in the future from cases of chromoblastomycosis should be a very simple problem.

Treatment. Chromoblastomycosis has been subjected to various methods of treatment with varying degrees of success. Up to the present time, no specific drug has been discovered against this dreadful malady. In incipient cases, however, the infection has often been successfully eradicated by surgical or electrotherapeutic methods. When the pathologic process is advanced, amputation of the affected extremity is the only hope for recovery. The iodides, copper used in different forms, and a few other drugs

have been more or less helpful in the hands of different investigators. Local treatment should be conducted along general principles.

ADDENDUM

The following Latin diagnosis is given in compliance with the International Rules of Botanical Nomenclature:

FONSECAEA PEDROSOI (Brumpt) Negroni, 1936, emend, variety PHIALOPHORICA.

Syn.: *Phialophora macrospora* Moore & Almeida. Ann. Missouri Bot. Gard. **23**: 543–552. 1936.

Phialophora verrucosa, A. Pedroso & J. M. Gomes. Bull. Soc. Med. Cir. São Paulo **3**: 254. 1920; Gomes, *ibid.* **3**: 42, 43. 1920; Ann. Paulistas Med. Cir. **11**: 53–61. 1920.

Acrotheca Pedrosoi Terra, Torres, da Fonseca & Area de Leao. Brasil Medico **2**: 363–368. 1922.

Morphologia essentialiter similis *Fonsecaea Pedrosoi typicus*, sed Phialophora sporulatio frequentissima, Acrotheca sporulatio rara et Hormodendrum sporulatio obsoletus.

SCHOOL OF TROPICAL MEDICINE,
SAN JUAN, P. R.

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POLYCHYTRIUM: A NEW CLADOCHYTRIACEOUS GENUS

LIBERO AJELLO[1]

(WITH 16 FIGURES)

This fungus was collected in decaying vegetable debris from a bog on the ridges of Bearfort Mountain, New Jersey, west of Greenwood Lake, and cultured in bleached sections of young corn leaves in the laboratories at Columbia University. It is characterized by a coarse rhizomycelium, polymorphic sporangia, and hyaline zoöspores with a well developed lunate opaque body and no prominent refractive globule. Inasmuch as it differs in several respects from any of the known genera and species of the Cladochytriaceae a new genus is hearby created for this chytrid. Because of its polycentric type of growth and aggregated sporangia the following names are proposed:

Polychytrium gen. nov.

Rhizomycelium intra- and extramatrical, extensive, coarse, branched, occasionally septate with rhizoids, conspicuous spindle organs or swellings lacking. Zoösporangia non-operculate, terminal and intercalary, variously shaped, spherical, clavate or pyriform. Zoöspores posteriorly uniflagellate, emerging fully formed in a globular mass and remaining quiescent for a few moments before swimming away.

Rhizomycelio intra- et extramatricali, extenso, crasso ramosque, interdum septato cum rhizoideis, neque tumores neque conspicua corpora fusiformia praestante. Zoosporangiis terminalibus et intercalaribus, variatim formatis, sphaericis, clavatis aut pyriformibus, neque operculatis. Zoosporis a posteriore uniflagellatis, maturis in globuloso cumulo emergentibus, aliquamdiu quiescentibus, postea enatantibus.

Polychytrium aggregatum sp. nov.

Rhizomycelium extensive, coarse, tenuous portion, apart from rhizoids, 2–12 μ in diameter, profusely branched, occasionally sep-

[1] The writer wishes to express his sincere appreciation to Professor J. S. Karling for helpful advice and criticism during the course of this study.

tate, hyaline at first, becoming yellowish-brown at maturity. Zoösporangia in aggregates of two or more, terminal and intercalary, non-apophysate, hyaline at first, becoming yellowish-brown at maturity, wall .7 μ thick; smooth to tuberculate; spherical, 14 × 29 μ; ovoid, ellipsoid, 12–20 × 22–40 μ; clavate, obclavate, 12–24 × 29–102 μ; pyriform, obpyriform, elongate, cylindrical, 8–25 × 17–75 μ; tubercles on sporangia up to 7 μ wide at the base and 5.5 μ in height; exit pore or tube varying in length, diameter 3.5 μ; proliferating, exit tube of secondary or tertiary sporangia often penetrating the primary sporangial wall. Zoöspores delimited within the sporangium, emerging and forming a motionless, spherical mass at the mouth of the exit pore; spherical 4.4–5.5 μ with a conspicuous, large, lunate opaque region, 1.5–2 × 3–3.5 μ, surrounded by several opaque granules, no conspicuous single refractive globule present; flagellum 24–29 μ long. Resting spores unknown or doubtful.

Fungus saprophyticus; rhizomycelio extenso, crasso, parte tenui (rhixoideis exclusis), 2–12 μ diametro, maxime ramoso, aliquando septato, hyalino primo, sed maturitate fulvoso. Zoosporangiis duobus aut pluribus aggregatis, terminalibus et intercalaribus, sine apophysate, hyalinis primo, maturitate fulvosis, pariete .7 μ crasso; polymorphis, levibus ad tuberculatis (tuberculo sporangii usque 7 μ lato ab infimo, 5.5 μ alto), sphaericis, 14 × 29 μ; ovoideis, ellipsoideis, 12–20 × 22–40 μ; clavatis, obclavatis, 12–24 × 29–102 μ; pyriformibus, elongatis, cylindriceis, 8–25 × 17–75 μ, porum exeuntem aut tubulum varia longitudine habentibus; proliferatis, tubulo exeunte secundorum aut tertiorum sporangiorum parietem primum penetrante. Zoosporis intra sporangium delimitatis, emergentibus et immotilem sphaericum cumulum orifice tubuli exeuntis formantibus; sphaericis, 4.4–5.5 μ, cum conspicua, permagna, lunata, opaca regione 1.5–2 × 3–3.5 μ, a compluribus opacis granulis circumdatis, neque conspicuo singulo globulo refractivo praeditis; flagello 24–29 μ longo. Sporis perdurantibus incompertis aut dubiis.

Saprophytic in decaying vegetation in bogs, Bearfort Mountain, Passaic County, New Jersey.

Polychytrium differs from the seven genera that at present comprise the family Cladochytriaceae in several respects. The sporangia of *Polychytrium* dehisce by the deliquescence of the tip of the exit-pore thus being clearly differentiated from the two operculate genera, *Nowakowskiella* and *Septochytrium*. *Amoebochytrium* is unique in having aflagellate zoöspores, *Polychytrium* and the other genera of the family having posteriorly uniflagellate spores. The zoöspores of *Catenaria* usually emerge singly from the zoösporangium and immediately swim away. Those of *Poly-*

chytrium, Cladochytrium, Nowakowskiella and *Septochytrium* emerge and form a more or less globular mass at the mouth of the exit tube. After a short quiescent period the mass breaks up and the spores swim away. The zoöspores of *Physocladia,* on the other hand, according to Sparrow (**1931**) behave quite differently from those of all the other genera in that they are confined in a thin but rigid, hyaline vesicle upon emerging from the sporangium. The rhizomycelium of *Polychytrium* is coarse and myceloid in character with no spindle organs or intercalary swellings. Rhizoids are not as numerous nor as extensively developed as those of various species of *Cladochytrium.* They are frequently difficult to distinguish, and offhand the rhizomycelium looks strikingly like the mycelium of members of the *Oömycetes* or Zygomycetes. In comparison to *Polychytrium* the absorbing system of the other genera of the Cladochytriaceae varies from the thick, almost cylindrical rhizomycelium as in *Catenaria,* which also lacks well-defined spindle organs to the extensively branched thallus of *Cladochytrium* in which intercalary swellings are numerous and well developed. *Physoderma* is markedly different from *Polychytrium,* for it has septate turbinate organs and its evanescent thin-walled sporangia arise on monocentric thalli.

DEVELOPMENT OF THE THALLUS

The living zoöspores of *P. aggregatum* are distinguished from most of the members of the family Cladochytriaceae by the lack of a conspicuous refringent globule and the presence of an opaque lunate body, 3–3.3 μ in diameter, which is usually bordered by several minute granules (FIG. 2). In only one other species of this family has such an opaque body been reported in living zoöspores. The zoöspores of *Catenaria sphaerocarpum* were described by Karling (**1938**) as containing a crescentic, opaque body, but its zoöspores also include a large refractive globule. Fixed and stained zoöspores of other chytrids and Phycomycetes have revealed crescentic bodies similar in appearance to the lunate structure observed in the living zoöspores of *P. aggregatum.* Karling (**1937**) found that the fixed zoöspores of *Cladochytrium replicatum* contained an extra-nuclear cap, and Hillegas's (**1940**) cytological investigation of *Endochytrium operculatum* also revealed such a

body in the zoöspores. Extra-nuclear caps have also been described in the zoöspores of *Coelomycidium Simulii, Rhizophidium beauchampi* and *Clavochytridium stomaphilum* by Debaisieux (**1920**), Hovasse (**1936**) and Cox (**1939**) respectively. Extra-nuclear caps are more prevalent and well developed in the Blastocladiales. In various species of *Blastocladia* and *Allomyces* Thaxter (**1896**), Barrett (**1912**), Kniep (**1929**), Cotner (**1930**) and Hatch (**1935**) found well developed and striking extra-nuclear caps. These structures are doubtless more widely distributed among the lower fungi than is generally supposed and their presence may or may not be specific and fundamentally significant in phylogeny. Aside then for the presence of an opaque, lunate body visible in the living zoöspores and the lack of a conspicuous refractive globule, the zoöspores of *P. aggregatum* resemble in form and activity those of other species in the family Cladochytriaceae. The zoöspore is medium in size (4.4–5.5 μ) (FIG. 2) and spherical in shape upon emergence from the sporangium. Since the diameter of the exit tube or pore is only approximately 3.5 μ the zoöspores become elongate while emerging (FIGS. 1*f* and 3). The zoöspores swim with the aid of a single, posteriorly attached flagellum 24–29 μ in length. Occasionally a globule of cytoplasm or a loop was observed at the tip of a zoöspore's flagellum (FIG. 4). Such zoöspores have been reported for *Synchytrium* by Curtis (**1921**), for *Macrochytrium* by Minden (**1923**) and by Berdan (**1941**) for *Cladochytrium*. These workers attributed this phenomenon to disintegration. Hillegas (**1940**) in studying *Endochytrium* believed it to probably be a developmental stage. After a motile period of varying length the zoöspores settle down and germinate.

Few stages of germination were observed, but in figure 5 a germinating zoöspore is shown from which a coarse, branched germ tube has developed. This tube grows in length and width and eventually forms the rhizomycelium. The thallus is hyaline when young but soon turns a yellowish-brown in color. It is coarse and profusely branched, with feebly branching rhizoids arising at various points (FIGS. 1*d* and 6). Except for the presence of rhizoids the tenous portion of the rhizomycelium, in which septa are occasionally formed (FIG. 1*j*), has the appearance of a coarse mycelium as in the higher, strictly mycelial fungi. Sporangia in various

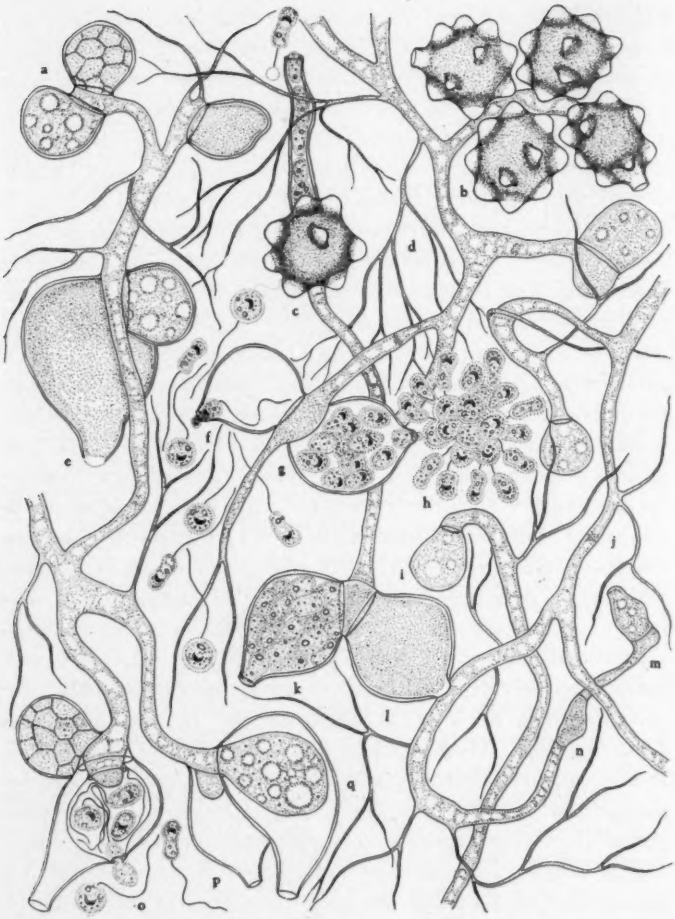
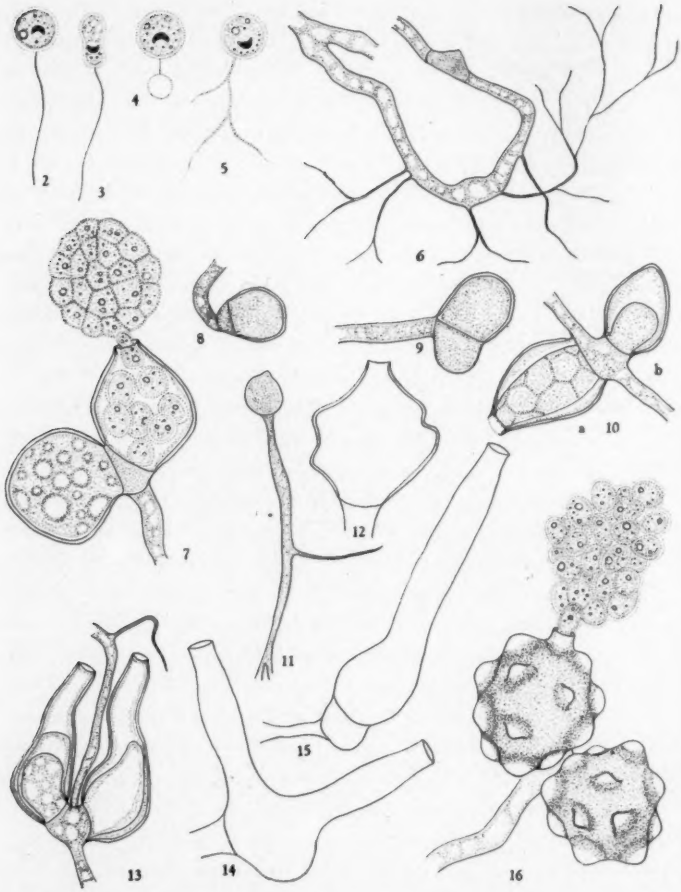


FIG. 1 *a-q*, habit sketch of the rhizomycelium of *Polychytrium aggregatum*. *a*, two ovoid zoosporangia in different stages of development, the upper sporangium undergoing cleavage; *b*, group of tuberculate zoosporangia; exit pore of two visible; *c*, solitary tuberculate sporangium with long exit tube; zoospores just beginning to emerge; *d*, group of branched rhizoids; *e*, pyriform sporangium with homogeneous cytoplasm; companion sporangium much smaller; *f*, zoospore being constricted while trying to emerge from the zoosporangium; *g*, sporangium discharging zoospores; *h*, zoospore mass breaking up; opaque lunate body and flagella visible; *i*, early stage in sporangial development; septa have cut off the sporangium from the thallus;

stages of development are shown in parts *i*, *m* and *n* of figure 1, which is a habit sketch of the rhizomycelium and also in figures 8, 9 and 11. With age the cytoplasm of the thallus tends to become vacuolate. Zoösporangia appear at various points at the apex and in intercalary positions (FIGS. 1*a* and *b*), and are delimited from the rhizomycelium by septa (FIG. 1*k* and *l*). As the specific name of the chytrid indicates, the zoösporangia rarely occur singly (FIG. 1*c*). In the incipient stages they often seem to arise singly, since one of the pair may develop at a faster rate than its companion (FIG. 1*n*), but in time the two sporangia become approximately equal in size (FIGS. 1*i*, *k*, *l*, *m*, 8 and 9). These sporangia undergo cleavage and form zoöspores either at the same time (FIG. 1*f* and *g*) or separately (FIG. 7). The tuberculate sporangia begin as swellings in the rhizomycelium, which increase in size and develop in the same manner as the smooth sporangia. As noted before, no conspicuous oil-like refractive globules are present in the developing zoösporangia as in the incipient sporangia of most chytrids. Before the zoöspores are delimited by cleavage the cytoplasm becomes homogeneous in appearance and contains scattered opaque granules (FIG. 1*e*). After cleavage the apical portion of the exit pore deliquesces and the zoöspores begin to emerge in the same manner as in *Cladochytrium, Nowakowskiella,* etc. (FIG. 1*k*). The first to emerge are surrounded by a viscous fluid-like substance which holds the zoöspores together at the mouth of the exit pore (FIG. 16). Such a slimy matrix appears to be common of all chytrid species of which the zoöspores remain clustered for a short time at the mouth of the exit pore. In *Polychytrium aggregatum* this zoöspore mass rounds up (FIG. 7) and remains motionless as more zoöspores slips into it. In a few moments the individuals making up the mass begin to move and glide over one another. It is at this time that the flagella can first be seen clearly,

companion sporangium not as yet developed; *j*, septum in the rhizomycelium; *k*, zoospores about to emerge from the sporangium; lunate opaque body and flagella not as yet visible; *l*, companion to sporangium *k* in an early stage of differentiation; *m*, young sporangium with accompanying sporangium just beginning to develop; *n*, incipient intercalary sporangium; *o*, 'secondary' sporangium whose exit pore has broken through the primary sporangial wall; a 'tertiary' sporangium is beginning to form within the 'secondary' one; *p* and *q*, stages in sporangial proliferation.



FIGS. 2–16. 2, zoospore showing opaque lunate body surrounded by granules; 3, zoospore elongated while emerging from sporangium; 4, zoospore with a loop at the tip of the flagellum; 5, germinating zoospore; 6, portion of the rhizomycelium showing an incipient intercalary zoosporangium and rhizoids; 7, pair of apical sporangia in different stages of development; 8, young zoosporangium; companion sporangium just beginning to develop beneath; 9, later stage in the development of companion sporangium; 10, proliferating zoosporangium; *a*, cleavage of 'secondary' zoosporangium; *b*, young 'secondary' sporangium; 11, early stage in development of an apical sporangium; 12, median optical view of a tuberculate zoosporangium showing the thickness of its wall; 13, pair of proliferating intercalary zoosporangia

and by the lashing about of the flagella the zoöspores emerge from the mass and swim away (FIG. 1*h*). The zoöspores which emerge from the sporangium after the mass breaks up have a clearly visible flagellum (FIG. 1*o*). The opaque lunate body of these zoöspores is also quite evident at this time in contrast to its faint appearance in the initial zoöspores which compose the mass at the mouth of the exit tube. In the latter zoöspores the lunate body becomes more visibly prominent at the time of the break up of the zoöspore mass (FIG. 1*h*).

The shape of the sporangia varies widely in *P. aggregatum,* and ranges usually from spherical, ovoid, to pyriform (FIG. 1*a* and *e*). Long cylindrical sporangia also occur (FIG. 15). Beside these variations in sporangial form, tuberculate sporangia may also be present on the same thallus with the smooth ones. These sporangia usually have 8 to 11 well developed tubercles upon their surface (FIGS. 1*b* and 16) which measure up to 7 μ at the base and 5.5 μ in height. In the incipient stages, the tuberculate sporangia are indistinguishable from the smooth ones and arise in the same manner either in an intercalary or apical position. As they develop further, however, the tubercles begin to form and at maturity they too become yellowish-brown in color. Whether or not these tuberculate sporangia should be considered as resting spores is not certain at present because their sporangial wall is not appreciably thicker than that of the smooth sporangia (FIG. 12). They are nevertheless, very similar in appearance to the resting spores of most cladochytriaceous species. Their exit tubes may be well developed (FIG. 1*c*) or commonly papillate, solitary (FIG. 1*k*) or in twos (FIG. 14).

Sporangial proliferation is quite common in this chytrid, with the secondary and tertiary sporangia being formed within the empty shell of the primary one (FIG. 13). Early stages in the formation of "secondary" sporangia are shown in figure 1*p* and *q*. The proliferating sporangia are formed by the ingrowth of the protoplasm from the rhizomycelium beneath as in most polycentric chytrids. The incipient "secondary" sporangium is at first hyaline

with 'secondary' and 'tertary' sporangia; 14, zoosporangium with two exit tubes; 15, cylindrical sporangium; 16, pair of tuberculate sporangia; upper one discharging zoospores.

and homogeneous (FIG. 10*b*) and undergoes the same stages of enlargement and differentiation as the primary sporangia (FIGS. 10*a* and 13). The exit papillum of the proliferating sporangium may be formed within the primary sporangium (FIG. 10*a*) or may push through the wall of the enclosing sporangium (FIG. 1*o*).

SUMMARY

Polychytrium aggregatum is a new, polycentric, saprophytic species of the family Cladochytriaceae which occurs in the decaying vegetation of bogs in the ridges of Bearfort Mountain, Passaic County, New Jersey. It has a coarse, richly branched rhizomycelium which becomes yellowish-brown at maturity, and lacks spindle organs or intercalary enlargements. The sporangia are smooth or tuberculate and produce spherical, posteriorly uniflagellate zoöspores which lack a conspicuous refractive globule but include a prominent opaque lunate body. The sporangia dehisce by the deliquescence of the tip of the exit tube or papilla. Dormant thick-walled resting spores have not been observed, but the irregular tuberculate yellowish-brown sporangia are strikingly similar to the resting spores of many Cladochytriaceous species. However, they produce zoöspores directly without going through a dormant period.

DEPARTMENT OF BOTANY,
COLUMBIA UNIVERSITY,
NEW YORK, N. Y.

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COCCIDIOIDOMYCOSIS [1]

C. W. EMMONS

(WITH 18 FIGURES)

Coccidioidomycosis was first studied and reported by Posadas (**30**) and Wernicke (**38**) in South America. Observing a resemblance of the fungus seen in tissue sections to certain Coccidia they described the condition as a new protozoan disease. In 1894 Rixford independently found the disease in California. Rixford and Gilchrist (**32**) later reported this case, described it as coccidioidal pseudotuberculosis, and named the organism *Coccidioides immitis*. In 1900 Ophuls and Moffitt (**29**), studying the third North American case, proved by culture that the etiological agent was a fungus. The original misconception of the nature of the microorganism does not invalidate the name and the fungus is properly referred to as *C. immitis* Rixford & Gilchrist 1896.

The disease occurs in two forms (**13, 14, 22, 23, 35**). One is a benign, acute, self-limited, respiratory infection; the second is a grave, chronic, generalized, progressive, granulomatous disease with a mortality rate of about 50 per cent. Both have been known for many years in the San Joaquin Valley of California and were believed to be two unrelated diseases until Gifford (**22, 23**) and Dickson (**13, 14, 15**) demonstrated that *C. immitis* is associated with both conditions. Dickson (**13**) at that time proposed the names primary and secondary or progressive coccidioidomycosis by which the two types of the disease are now generally known.

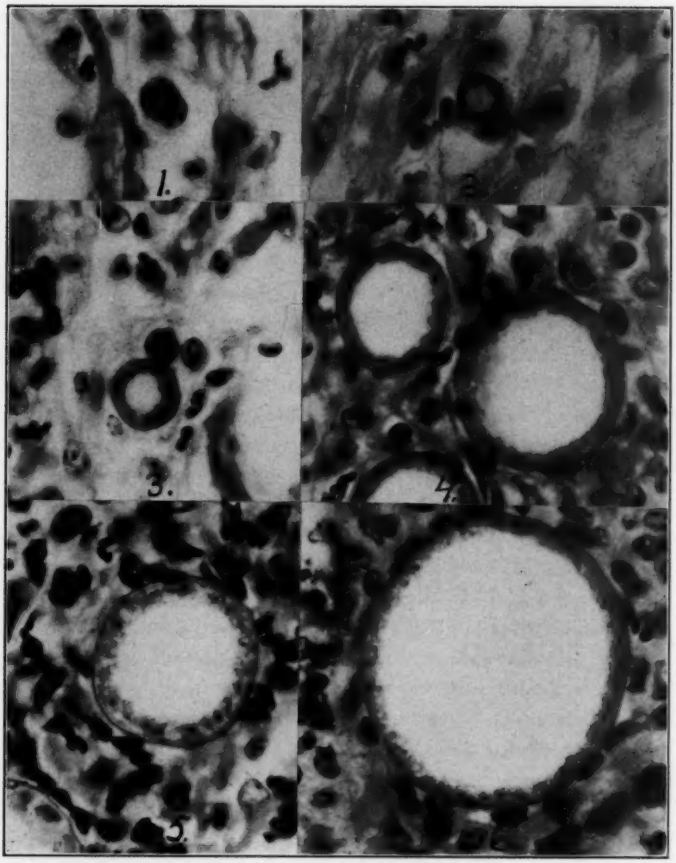
The first of these, primary coccidioidomycosis, varies widely in severity. It probably occurs in many individuals in a form so mild as to be unrecognized. The severity of recognized cases may correspond to that of a common cold. More severe cases may resemble influenza and may be accompanied by high fever, pneumonia, and formation of pulmonary cavities. In perhaps 5 per cent of those infected erythema nodosum may be expected and

[1] Division of Infectious Diseases, National Institute of Health.

the disease is then recognized clinically as Valley fever, San Joaquin fever, desert rheumatism, "the bumps," etc. Irrespective of the severity of the primary disease, spontaneous recovery usually follows. Epidemiological studies (**34**) seem to indicate that it progresses to the secondary type only in exceptional cases. Reinfection appears to be infrequent.

Progressive or secondary coccidioidomycosis (**6, 7, 32, 33**) (coccidioidal granuloma) is manifested by cutaneous, subcutaneous, visceral, and osseous lesions. It often resembles tuberculosis so closely that a differential diagnosis can be made only by the laboratory demonstration of the fungus. It was once believed to be invariably fatal but milder and arrested or healed cases are now recognized. It is not definitely known whether progressive coccidioidomycosis results from a reinfection or whether it is a reactivation of a latent or temporarily arrested lesion of the primary disease. The fungus has been recovered in culture from arrested and partially cacified lesions in individuals dying of other causes (**5, 8, 15**). Coccidioidomycosis occurs also in cattle, sheep, dogs, and rodents (**10, 11, 4, 20, 24, 18**).

The disease is of frequent occurrence in the San Joaquin Valley and probably over large areas of the arid Southwest. It is rarely seen or is unknown elsewhere. Strangely enough, it appears to be rare in South America where it was first seen. Most of the reported cases from this area were paracoccidioidal granuloma (**12, 25**), a disease differing in clinical characteristics and etiology. Coccidioidal granuloma is a reportable disease in California, and by June, 1939, 578 cases and 278 deaths had been reported (**21**). It is more difficult to determine how frequently primary coccidioidomycosis occurs. Individuals with the respiratory type of infection do not always raise sputum and an attempt to demonstrate the presence of the fungus may fail even in severe cases which are clinically typical of coccidioidomycosis. In an "epidemic" of seven cases probably infected from a common exposure, the fungus was demonstrated in only three of the seven (**31**). In Smith's (**34**) epidemiological study of 432 cases of Valley fever occurring in the San Joaquin Valley during a period of 17 months, Coccidioides was recovered from only 22 per cent of the patients. Smith estimated that only about 5 per cent of those who have had



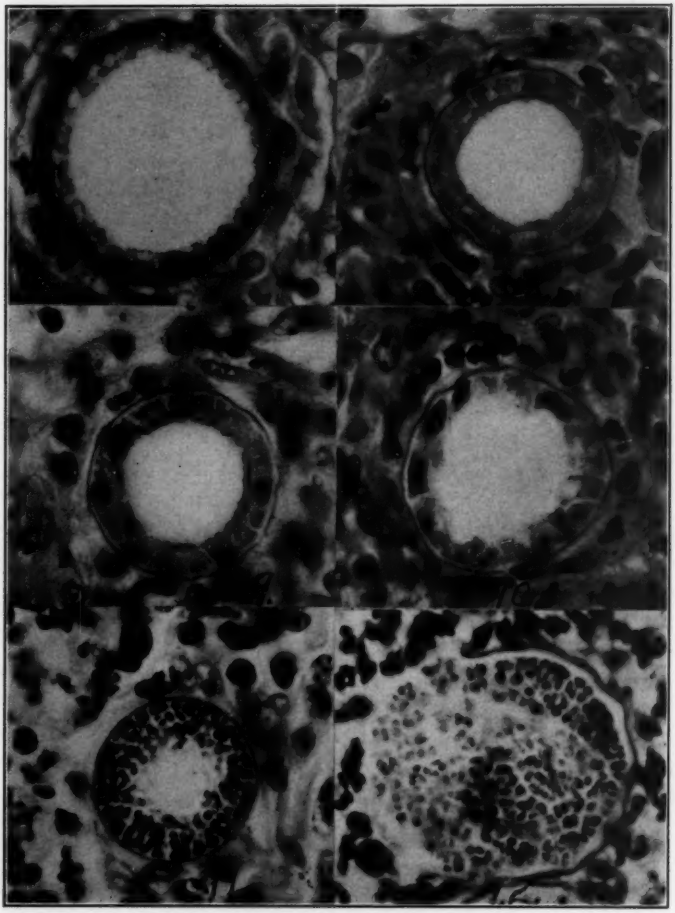
FIGS. 1–6. *Coccidioides immitis.*

the infection presented clinical symptoms sufficiently severe and well defined to allow a clinical diagnosis to be made. This estimate was based on information obtained by the use of a skin test similar to the tuberculin test.

The testing material, coccidioidin (**19**), is prepared by allowing strains of the fungus to grow for two months in a synthetic broth. The broth is then filtered and the sterile filtrate is diluted and 0.1 c.c. is injected intradermally. An area of erythema and edema

appears around the site of injection in sensitized individuals. The reaction is read in 24 and 48 hours after injection. It gradually disappears. The assumption that the test is specific and that a positive reaction indicates a previous infection with the fungus is supported by several lines of evidence (**36**, **34**). Skin sensitivity suddenly develops in man a few days or a few weeks after attacks of primary coccidioidomycosis. A recent infection usually gives rise to a more severe reaction than an infection incurred several years previously. Individuals with secondary coccidioidomycosis react less strongly than those with the primary type, and in the terminal stages may fail to react. A cross reaction with tuberculosis and other diseases has not been clearly demonstrated. Experimentally infected guinea pigs acquire a sensitivity to the intradermal injection of coccidioidin. The coincidence of a high percentage of positive reactions in the residents of an endemic area and their rarity or absence elsewhere is also noteworthy. There is not complete agreement on the correct interpretation of the coccidioidin skin test (**36**), and the preparation of coccidioidin of reproducible potency still presents difficulties, but the test is the best available method of determining the prevalence of past Coccidioides infections.

Some of the recent data on the prevalence of skin sensitivity to coccidioidin in residents of endemic areas and its rarity or absence in other populations have been brought together by Farness (**21**). From 16 per cent to 90 per cent of the individuals in certain groups within endemic areas react. The highest percentage of reactors was reported by Aronson, et al. (**1**, **2**), who found that a very high percentage of the Indian school children on the San Carlos, Pima, and Papago Indian reservations in southern Arizona reacted to coccidioidin. In spite of the prevalence of positive skin reactions neither primary nor progressive coccidioidomycosis is commonly seen in these groups. Suspected cases were seen on the reservations, and histories of earlier suspected cases were obtained, but none of these were proved. It seems probable that the disease is prevalent in these areas and that most of the adult residents have at some time been infected, in most cases during early childhood. It is not yet apparent why the disease is not often seen and recognized on these Indian reservations. It is probable that



FIGS. 7–12. *Coccidioides immitis.*

in the comparatively stable populations of these areas it is an unrecognized mild disease of early childhood. In the San Joaquin Valley it is seen as a more severe disease among migrant or newly resident adults or children of school age (**22, 34**).

So far as we know, coccidioidomycosis is not transmitted directly from man to man (**34**). The parasitic phase of the fungus

which occurs in human pus and sputum is infectious when experimentally injected into animals, but apparently is not effective in the natural direct transmission of the disease. Spores from the saprophytic growth phase of the fungus are also infectious. Epidemiological studies (**16, 34, 35**), accidental laboratory infections (**14, 17, 34**), and experimental infection of guinea pigs by inhalation (**9**) make it seem probable that man is ordinarily infected by inhalation of air-borne spores of the fungus. There is a remarkable association of cases of the disease and previous exposure to dust storms or occupational exposure to agricultural dust (**17**). It is generally assumed that the spores of the saprophytic growth phase of the fungus are present in such dust and that the fungus was growing in the soil from which the dust arose. Additional evidence for both assumptions is desirable. In spite of many attempts to isolate Coccidioides from soil, success has been reported only three times. The first isolation was from soil taken near the sleeping quarters of a Delano ranch where there were four cases of progressive coccidioidomycosis (**37**). Contamination of this soil by pus and sputum from the patients may have accounted for the success in this isolation. The second instance (**35**) was in San Benito County, California, and the details of this isolation have not yet been published. Both these sites were within the known endemic area of Southern California.

Recently five isolations of *Coccidioides immitis* were made from desert soil collected at distances up to four miles from human habitations in the vicinity of San Carlos, Arizona (**18**). The fungus was also isolated for the first time from rodents (**18**). These isolations are further noteworthy because they demonstrated the presence of the fungus in that area before the occurrence of the disease was conclusively demonstrated. No clinical case of coccidioidomycosis in man has yet been proved on the San Carlos Indian Reservation, although the disease undoubtedly occurs there.

Coccidioides appears in animal tissues (**15, 28, 29, 32, 39**) only as spherical cells which vary in diameter from spores of one or two microns to mature sporangia 30–60 μ in diameter. This parasitic growth phase may develop *in vitro* under certain conditions (**27, 3**). On ordinary artificial culture media, it is a Hyphomycete reproducing by conidia or oidia (**29, 15**). Each of these forms

will be considered in some detail with descriptions of certain cytological and morphological details not hitherto described.

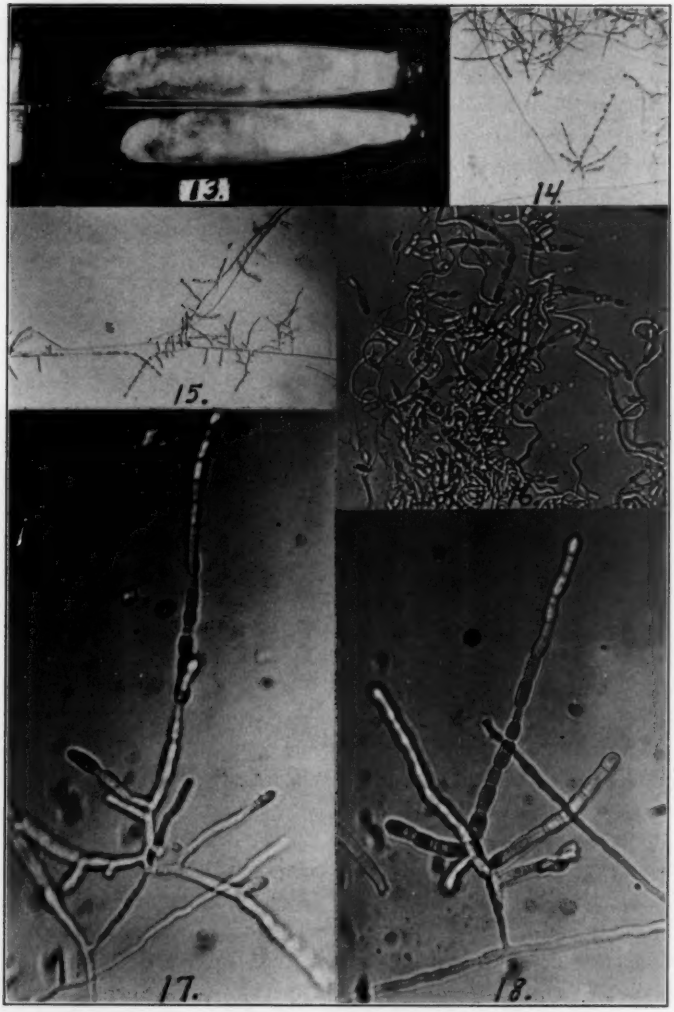
The newly disseminated spores in animal tissue or pus are often difficult to demonstrate. They may be intracellular (FIG. 1) or intercellular. As they increase in size, they retain the spherical shape (FIGS. 1–6). Budding never occurs. A central vacuole is apparent in the early stages of enlargement (FIG. 2). In older individuals this central vacuole occupies most of the cell, the stainable cytoplasm being distributed in a thin peripheral layer (FIG. 6). As the cell assumes the functions of a sporangium the amount of peripheral stainable cytoplasm increases and becomes vacuolate (FIG. 7). The small vacuoles in this peripheral layer probably determine the location of cleavage planes which now form radially (FIG. 8). Cell walls are laid down along these cleavage planes and delimit an indefinite number of large protospores (FIGS. 9, 10). The progressive formation of additional septa in both radial and tangential planes subdivides the protospores into spores with a diameter in most cases of 1–3 μ (FIG. 11). This process resembles the method of sporangiospore formation which Harper (**24a**) described for the Phycomycetes. The mature sporangium is filled with these spores. Its wall then breaks, allowing them to pass into the surrounding host tissues (FIG. 12). This simple cycle is repeated and constitutes the sole parasitic growth phase of the fungus. The general features of this developmental cycle have been well illustrated in numerous papers. Numerous variations such as the occasional formation of larger spores in certain fungus strains or in certain host tissues, and the precocious development of spores while still within the sporangium have also been described.

Apparently no attention has been given to the nuclear condition, probably because the nuclei are not readily demonstrated in the usual histological preparations. The preparations illustrated herewith were fixed with a modification of Bouin's fixative and stained with Haidenhain's iron alum hematoxylin. The spores probably contain a single nucleus in most instances (FIG. 12), but a multinucleate condition is early established in the developing cell (FIGS. 2, 3). These nuclei are typical of fungus nuclei, having a thin nuclear membrane and a single nucleolus. As the cell increases

in size, an increasing number of nuclei appear in the peripheral layer of cytoplasm (FIG. 6). These are not separated by cell walls, but are scattered, the spatial relationships being more apparent in slab sections of cells than in the median sections illustrated. After the process of spore delilimation is initiated, the protoplasm is divided by the newly formed walls into multinucleate protospores in the manner previously described (FIG. 10). The spores which result from the further subdivision of the protoplasm appear to be uninucleate in most cases (FIG. 12). Mitotic figures were not found in these preparations.

The saprophytic growth phase on artificial culture media is that of a mold. Growth is rapid. The colony may be glabrous at first, but aerial hyphae are usually formed in abundance (FIG. 13), at least in the center and in a peripheral zone. The general aspect is that of a rather coarse growth but actually most of the hyphae are unusually delicate. The color of the colony is gray or brownish white.

It is commonly stated that the aerial hyphae break up into chlamydospores. This is the impression one gains from the examination of an old culture (FIG. 16). Long chains of spores or fragments of chains are conspicuous but the details of spore production cannot be observed in such preparations. If a five day old culture is carefully examined, it is apparent that the spores are not merely the segmented fragments of the aerial vegetative hyphae. They are borne on well differentiated conidiophores (FIGS. 14, 15, 17, 18). These arise as specialized side branches which are almost twice the diameter of the vegetative hyphae and may be simple (FIG. 15) or branched (FIGS. 14, 17, 18). Septa are formed at frequent intervals on the terminal portions of these conidiophores. Alternate cells, after being thus delimited, increase in size and turgidity and in thickness of the wall (FIG. 18). The intervening cells cease development and gradually lose any demonstrable cytoplasm. The walls persist and hold the spores together in chains (FIG. 16). These chains of mature spores separated by dead or empty cells are familiar from numerous published photomicrographs and drawings. They resemble and are usually called chlamydospores. From a consideration of the manner in which they are borne, it is suggested that they should instead be designated conidia or oidia.



FIGS. 13–18. *Coccidioides immitis.*

Spirally coiled hyphae similar to those found in certain strains of dermatophytes are frequently present (FIG. 16). Baker and Mrak (3) have recently described the development in old agar

cultures of sporangia similar to those which characterize the parasitic phase of growth.

SUMMARY

Coccidioidomycosis is a mycotic disease which occurs in a benign primary form and as a grave progressive disease. It has a limited geographic distribution. The prevalence of positive coccidioidin skin tests in endemic areas indicates that a high percentage of the residents of such areas have at some time been infected. The disease is most apparent in migrant or newly resident adults. The parasitic growth form of the fungus is infectious but appears to be ineffective in the direct natural transmission of the disease. There is an apparent association between exposure to dust storms or occupational exposure to agricultural dust and subsequent infection. It is generally assumed that spores of the saprophytic growth form are present in such dust. The fungus has been isolated from soil in three areas and from cattle, sheep, dogs, and rodents. The multinucleate condition of the fungus, sporangiospore formation in the parasitic growth form, and the development of the conidiophores and conidia of the saprophytic growth form are described.

NATIONAL INST. OF HEALTH,
WASHINGTON, D. C.

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EXPLANATION OF FIGURES

Coccidioides immitis. Magnification of Figs. 1–12 about 600 ×. Fig. 1, Young vegetative cell within a phagocytic cell. The multinucleate condition is already established. Fig. 2, Young vegetative cell showing several nuclei and a large vacuole. Fig. 3, Two young vegetative cells. Fig. 4, Increase in size of cell and in number of nuclei. Fig. 5, Vacuolate condition of peripheral layer of protoplasm. Fig. 6, Vegetative cell has reached mature size. Fig. 7, Increase in amount of protoplasm in young sporangium and appearance of vacuoles which precedes cleavage. Fig. 8, Cleavage planes and beginning of wall formation. Figs. 9 and 10, Formation of multinucleate protospores. Fig. 11, Subdivision of protospores by formation of septa in radial and tangential planes to form sporangiospores. Fig. 12, Rupture of sporangial wall. Fig. 13, Cultures one month old on acid dextrose agar. Fig. 14, Low power view of branched conidiophore. Fig. 15, Low power view of simple and branched conidiophores. Fig. 16, Spirals and chains of conidia from an old culture. Note intervening empty cells. Figs. 17 and 18. Branched conidiophores showing some details of formation of conidia. × about 600.

REVISIONARY STUDIES IN THE CORYNELIACEAE

HARRY MORTON FITZPATRICK

(WITH 43 FIGURES)

This paper, inclusive of a second part to appear in the next number of MYCOLOGIA, constitutes a revision of the writer's earlier monographic treatment of the Coryneliaceae, published in this journal over twenty years ago.[1] In the intervening period a considerable amount of additional material has been studied, some new and hitherto misplaced species have been incorporated, and changed viewpoints on essential features of morphology have resulted in altered conceptions of generic limits.

The family Coryneliaceaè was established by Saccardo[2] to embrace the two genera *Corynelia* Acharius and *Tripospora* Sacc. Later he[3] included also *Coryneliella* Hariot and Karsten. In 1897, in Engler und Prantl's Die Natürlichen Pflanzenfamilien, the family, consisting of these three monotypic genera, was placed by Lindau in the Sphaeriales alongside the Cucurbitariaceae. In the writer's taxonomic arrangement of the group the four genera *Corynelia, Tripospora, Sorica,* and *Caliciopsis* were recognized. The genus *Coryneliella,* known only from the type collection and clearly not a member of the family, was excluded. The paper constituted the first serious effort to provide adequate descriptions and separations of the species, and was based on the study of many more collections of material than had previously been assembled in any herbarium. The dehiscence of the ascocarp in *Corynelia* by definite apical cleavage, which had not been noted by earlier workers, was described and figured for the first time. The absence of a true ostiolum in all members of the family was emphasized, and relationship with the Perisporiaceae was suggested.

[1] Mycologia **12**: 206–267. *fig. 1–49.* 1920.
[2] Syll. Fung. **9**: 1073. 1891.
[3] Syll. Fung. **11**: 385. 1895.

In 1912 Arnaud,[4] who regards the fruit-body in *Corynelia* and related fungi as a true apothecium, had placed the known species in the Caliciaceae. Ten years after the publication of our monograph, he [5] presented a revision of his earlier paper, reiterating his belief that the fructification is discomycetous, and recognizing the Coryneliées as a subdivision of the Caliciaceae. His viewpoint arose largely from placing undue emphasis on the superficial resemblance of *Caliciopsis* to *Calicium*. In both genera the ascocarp is stipitate, and in both the ascospores extrude at its apex forming a pulverulent plug or knob. The fruit-body in the Coryneliaceae is, nevertheless, certainly not an apothecium. Though it opens widely at maturity, the arrangement of the asci is typically pyrenomycetous. They do not form a palisade-like hymenium. Instead they are fasciculate, and stand at various heights on extremely slender, long stalks. Avoidance of the term apothecium in our earlier paper led naturally, at that time, to the use of perithecium. It is now clear, however, in the light of later research on the ontogeny of the ascocarp in the loculate, stromatic series of higher Ascomycetes, that the fructification in the Coryneliaceae cannot properly be designated a perithecium. There is no perithecial wall, and the ascigerous cavity results from lysigenous action in the tissue of the stromatic lobe above the developing asci. Paraphyses and a true ostiolum are lacking, and the ascocarp is definitely dothideaceous in type. These points were demonstrated conclusively in our laboratory by Helene McCormack [6] in a study of the development of the fruit-body of *Caliciopsis pinea*, and corroborative evidence has been obtained by us in *Corynelia uberata*. It seems best at present to be content with the more general term ascocarp. Its use for the whole lobe containing the ascigerous locule has been adopted for the sake of uniformity in terminology throughout the paper, but is admittedly somewhat open to criticism in that the limits of stroma and ascocarp are only vaguely indicated. This is especially evident in *Caliciopsis pinea* and similar forms in which the locule occupies only a small part of a long, columnar projection of the pulvinate stroma.

[4] Ann. Ecole Nat. Agric. Montpellier, n.s. **12**: 24–49. 1912.
[5] Annales des Epiphyties **16**: 235–302. 1930.
[6] Mycologia **28**: 188. 1936.

The Coryneliaceae approach the Perisporiaceae in the form and nature of the ascocarp, and, except in *Corynelia,* in the type of its dehiscence. They differ in that the mycelium is endophytic instead of superficial, and in that the stroma is erumpent. Dehiscence in *Corynelia* is by apical cleavage, the tip of the ascocarp opening widely, usually along a single transverse suture. Though this recalls the situation in the Hysteriales, that group, on account of difference in form and arrangement of the asci, can scarcely be regarded as close. Though the interrelationships of the various groups of loculate Ascomycetes remain somewhat obscure, there is ample justification for retention of the family Coryneliaceae among them.

Inclusion, within the limits of the single family, of genera characterized by two wholly different types of dehiscence emphasizes our conviction that the species embraced are nonetheless closely related. Indeed, in our earlier paper the genus *Corynelia* included species representative of both types of dehiscence. Though we now remove from that genus the species in which dehiscence is by apical perforation, it is scarcely possible to transfer them to a separate family. In one of them, *C. fructicola,* the ascospores resemble the highly specialized spores of *Corynelia* far too closely to leave doubt as to the nearness of the relationship. Also, the fact that the species of three of the five genera incorporated in the family occur only on *Podocarpus* is regarded as significant.

The accessory fruit-bodies, occurring in various species and regarded by us earlier as pycnidia, are here termed spermogonia. This change is based largely on results obtained by Ray [7] in *Caliciopsis pinea.* Following artificial spermatization, he obtained ascocarps in pure culture on agar. Though similar work on other species has not been undertaken, the pycnidium-like body is probably uniform in character throughout the group.

CORYNELIACEAE Sacc. Syll. Fung. **9**: 1073. 1891.

Corynelieae Sacc. in Berlese & Voglino, Addit. Syll. Fung. 193. 1886.

[7] Mycologia **28**: 207. 1936.

Coryneliales Seaver & Chardon, Sci. Surv. Porto Rico & Virgin Islands, N. Y. Acad. Sci. 3[1]: 40. 1926.

Mycelium endophytic, in most cases parasitic; stromata formed within the host, early erumpent as coriaceous to carbonaceous, sharply demarcated, small, black cushions; surface of stroma soon putting out hemispherical to conical lobes which undergo further, vertical elongation and mature into spermogonia or ascocarps; spermogonia usually preceding the ascocarps, in some species lying among them, in others developed on separate stromata; mature spermogonium sessile to short-stipitate, with a minute, apical perforation; spermatia unicellular, elongate, hyaline to yellowish; ascigerous lobes undergoing considerably greater elongation than the spermogonial, in some species lengthening into slender, cylindrical columns; the entire lobe, regardless of its extent or form, termed here the ascocarp; ascocarp dothideaceous in type, lacking a true perithecial wall, and forming an ascigerous locule by lysigenous action in the stromatic tissue above the developing asci; location of the locule in the ascocarp varying in different species from basal to apical; apex of ascocarp rounded and undifferentiated, or definitely and variously lobed, never possessing a true ostiolum, in dehiscence opening widely by a single transverse cleft or several radiating ones, or perforated and dilated to funnel-form by the pressure of extruding ascospores; asci (p. sp.) ovate to clavate, with thin, evanescent walls and extremely long, delicate stalks, chiefly 8-spored; paraphyses and paraphysoids lacking; ascospores crowded, inordinate, unicellular, various in shape, smooth or echinulate, brown to hyaline, when very young polygonal from mutual pressure and with characteristically refractive centers.

KEY TO GENERA OF CORYNELIACEAE

A. Ascocarp apically dehiscent by a single, deep transverse cleft or several radiating ones; ascospores large, spherical, echinulate, thick-walled, and provided with prominent germ-pores .....1. *Corynelia*

B. Dehiscence not by cleft; ascocarp at maturity apically perforated and dilated to funnel-form by the extruding ascospores; the spores filling the funnel to overflowing, and giving it the aspect of a pulverulent, subglobose knob or convex disc; finally, following spore dissemination, the inner surface of the funnel exposed to view.

1. Four stout, radiating lobes giving the ascospore the form of a caltrop .....2. *Tripospora*

2. Ascospores spherical to ellipsoidal or subfusiform.

a. Ascospores closely resembling those of *Corynelia*, but less uniformly spherical and exhibiting greater variation in size.
3. *Coryneliospora*

b. Ascospores smooth.
(1) Ascospores large, with extremely thick walls; ascus 2-spored; species occurring only on *Podocarpus* .....4. *Lagenulopsis*
(2) Ascospores much smaller, with thin walls; ascus 8-spored; species not occurring on *Podocarpus* .....5. *Caliciopsis*

1. CORYNELIA Acharius ex Fries, Syst. Myc. 2: 534. 1823. (Obs. Myc. 2^2: 343. 1818.)

TYPE SPECIES, *Corynelia uberata* Fries.

Stromata rounded to slightly elongate, scattered to crowded, sometimes confluent, usually hypophyllous, not uncommonly amphigenous, caulicolous or fructicolous; all species parasitic on *Podocarpus* only; ascocarps usually covering the stroma more or less completely, but sometimes formed only at its margin as a single row of nearly horizontal, radiating individuals bordering a prominent, central cushion, in all cases appearing to be seated on the stroma, but actually with their bases somewhat buried in it, clearly not stipitate; mature ascocarp vertically elongate, composed usually of a rounded subconical to subcylindrical basal portion and a more or less broadly clavate upper part; the two usually tapering upward and downward respectively to provide a somewhat narrowed or constricted middle zone, thus giving the ascocarp a more or less dumb-bell shape; the whole interior of the ascocarp at maturity constituting an ascigerous locule, the lower portion containing the young asci, the upper part filled with more mature asci and free ascospores; the apex of the ascocarp differing in external aspect in the several species, in some definitely and characteristically lobed; dehiscence usually along a single, prominent, transverse, terminal groove, a deep cleft being formed which opens widely as a definite mouth; in some cases similar apical rupture accomplished by splitting along several radiating grooves, the resultant mouth bordered by three or more pointed, recurved lobes; ascus 1–8-spored; ascospores spherical, brown, thick-walled, and echinulate; the wall composed of a thin exospore and a thick endospore, and provided with a number of germ-pores which appear as lighter-colored, circular areas.

In the writer's earlier monograph, nine species were recognized. In two of these, *C. fructicola* and *C. bispora,* dehiscence is by apical perforation rather than by cleavage. These are now removed and made the types of two new genera, *Corynelíospora* and *Lagenulopsis* respectively. The other seven are retained in *Corynelia.* No additional species have meanwhile been added, but the study of

more favorable material has altered our conception of certain features, especially in *C. tropica* and *C. brasiliensis.* Our earlier diagnoses of all the species are here rewritten in abbreviated form in the light of improved knowledge and an altered terminology. As the members of the genus occur in widely separated and often inaccessible regions in lands remote from the mycological centers of the Northern Hemisphere, few students of the fungi have collected them, and few of even the larger herbaria contain more than a meager representation.

KEY TO SPECIES OF CORYNELIA

A. Apex of fully formed ascocarp not definitely lobed; dehiscence occurring along a shallow transverse groove, which crosses the apex but does not extend far down the sides; ascus 8-spored.
   1. Mature ascocarp definitely dumb-bell-shaped, and often bent or inequilateral; the apical portion finally of characteristically shaggy aspect ..... 1. *C. uberata*
   2. Ascocarp short-turbinate, with a smooth, rounded apex. ..... 2. *C. nipponensis*
   3. Ascocarp usually barrel-shaped or short-cylindrical; the sides marked by longitudinal ridges; the apex sometimes laterally compressed to form an indefinite beak ..... 3. *C. tropica*

B. Apex of fully formed ascocarp definitely lobed; dehiscence occurring between the lobes along rather deep grooves which extend far down the sides.
   1. Asci chiefly 8-spored; fewer-spored asci lacking or rare.
      a. Apex of ascocarp typically trilobed ..... 4. *C. oreophila*
      b. Apex of ascocarp typically bilobed ..... 5. *C. brasiliensis*
   2. Asci chiefly 3-spored; eight-spored asci lacking or rare.
      a. Apex of ascocarp typically trilobed ..... 6. *C. jamaicensis*
      b. Apex of ascocarp typically bilobed ..... 7. *C. portoricensis*

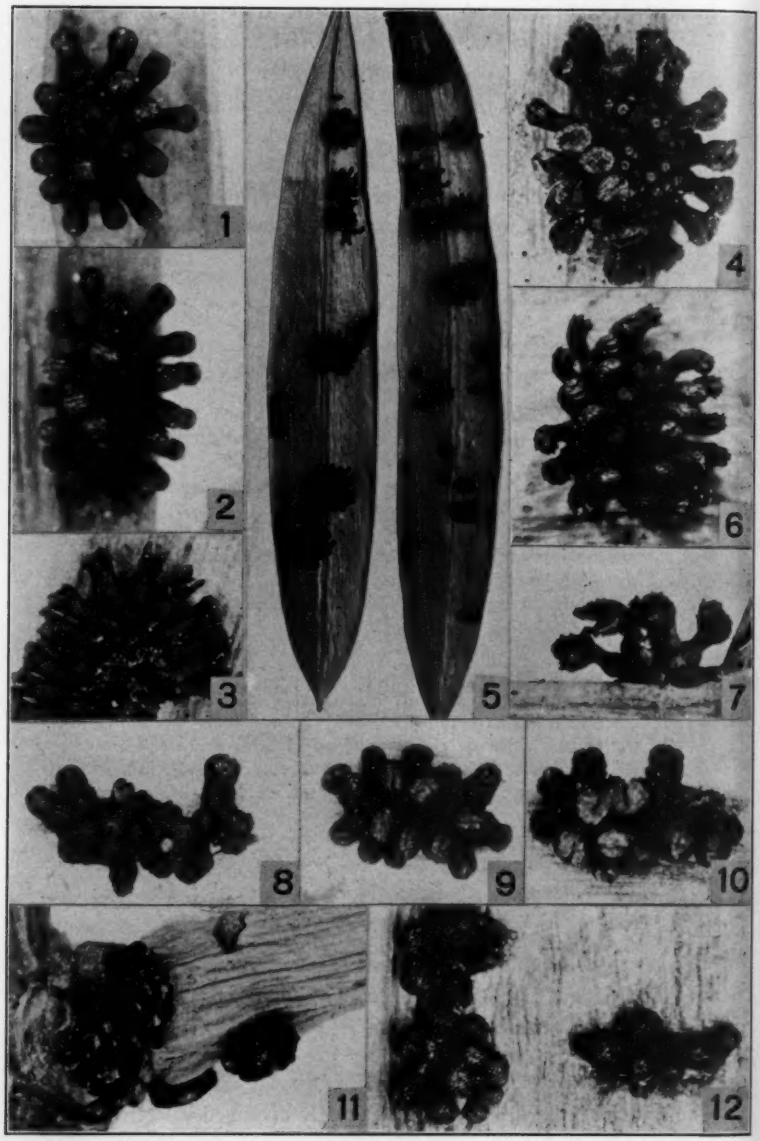
1. CORYNELIA UBERATA Fries, Syst. Myc. **2**: 535. 1823. (Obs. Myc. **2**^{2}: 343. 1818.)

Corynelia clavata (L.) Sacc., Nuovo Giorn. Bot. Ital. **21**: 312. 1889.

TYPE: specimen in herbarium of Fries at Upsala, Sweden, labeled by him *"Corynelia uberata* Fr. Cap. B. sp. Dedit Acharius, Exiguum at characterist."

(FIG. 1–7)

Stroma bearing a crowded, irregularly arranged cluster of ascocarps with occasional, smaller, spermogonia interspersed among



FIGS. 1–12.

them; young ascocarp conical, tapering from a dull, minutely roughened base to a smooth, shiny apex, with elongation becoming apically enlarged to form a broad, clavate beak; mature ascocarp approximately 1 mm. in length, strikingly dumb-bell-shaped, often curved or bent at the narrowed middle zone to give an inequilateral aspect; the swollen, terminal portion finally compressed laterally to form a transverse, apical ridge marked by a line or shallow furrow along which dehiscence occurs; paralleling this line on both sides, the apex commonly cut by one to several secondary furrows; the intervening ridges tending to break up into scales which give a pronouncedly shaggy aspect not present in other species; line of dehiscence merely crossing the apex of the swollen terminal beak, not continued far down its sides; in dehiscence the two lips often spreading far apart, exposing the lighter colored inner surface of the wall; asci (p. sp.) 34–44 × 20–26 μ, 8-spored; ascospores 9–14 μ (mostly 12) in diameter; spermogonium conical to flask-shaped, tapering to a short, prominently perforate beak; spermatia 5–7 × 2 μ.

We have studied more collections of this species than of any other, most of them having been made in South Africa, the region from which the species was originally described. We have also seen specimens from equatorial East Africa, Japan, and Australia. A single, fragmentary specimen from the Philippine Islands, cited in our earlier paper, is now regarded as doubtful. Though Cooke [8] records the species from New Zealand and figures ascocarps having

[8] Handbook of Australian Fungi, p. 318, *fig. 242.* 1892.

FIGS. 1–7. *Corynelia uberata.* 1, a cluster of ascocarps radiating from a stroma on a leaf of *Podocarpus spinulosa* from Australia, × 11. 2, another cluster from the same collection, with plane of focus adjusted to show the transverse apical furrows, the central one of which marks the line of dehiscence, × 11. 3, another cluster collected at the same place at a later date, showing apical dehiscence by a single transverse cleft, × 11. 4, a cluster from the same collection, showing the occasional extreme enlargement of the apical portions; spermogonia also present, × 11. 5, two leaves of *P. Thunbergii* from South Africa, bearing clusters of ascocarps, × 25. 6, one of these clusters showing deep apical furrows and pronounced tendency toward curvature, × 11. 7, another cluster of the African material, showing the occasional extreme enlargement of the apical portion of the ascocarp, × 11. FIGS. 8–12. *C. tropica.* 8, 9, clusters of ascocarps, from New Zealand, × 11. 10, dehiscent ascocarps from South America, × 11. 11, a twig of *P. totara* from New Zealand, with dense clusters of spermogonia, × 11. 12, clusters of immature ascocarps from the Philippine Islands, showing the apical dimple, × 11.

a more or less dumb-bell shape, our specimens from there are all referable to *C. tropica*. Hennings[9] reported *C. uberata* from Australia in 1903, but we had not seen material from there until 1935, when Lilian Fraser sent us collections from New South Wales showing the species in all stages of development. Photographs of some of her specimens are shown in figures 1–4 for comparison with the South African material, illustrated in figures 5–7. The ascocarp varies considerably in form in different collections. In some specimens its swollen, apical portion is considerably larger than the basal. The dumb-bell shape and the shaggy appearance of the apex are, however, sufficiently characteristic of *C. uberata* to render confusion with other species unlikely.

2. CORYNELIA NIPPONENSIS Fitzp. Mycologia **12**: 253. 1920.

TYPE: material of *Podocarpus macrophylla* Don., collected in Japan, was received at the Royal Botanic Gardens at Kew, January, 1893, from the Science College of the Tokyo Imperial University. George Massee noticed material of *Corynelia* on some of the leaves and transferred them to the cryptogamic herbarium. A part of the specimen was deposited at the New York Botanical Garden. The writer studied it there, and later received a portion of the material from Kew for comparsion. The species, *C. nipponensis*, was then erected on this single collection. Fifty years have now passed since the fungus was collected and twenty-two since the species was described. Meanwhile, no other material has been found. In the original diagnosis of *C. nipponensis* emphasis is placed on the turbinate shape of the ascocarp. The possibility that this represents merely a variation within the limits of *C. uberata* has been considered, but until material of intermediate character is found, it seems best to retain *C. nipponensis* as a distinct species. Although the ascocarps are admittedly immature, it should be emphasized that much young material of *C. uberata* has been examined without the discovery in any collection of characteristically turbinate individuals.

[9] Hedwigia **42**: 73. 1903.

(FIG. 27, 28)

Stroma sometimes circular, but more commonly elongated at right angles to the long axis of the leaf, erumpent through a transverse slit, bearing over the exposed surface a compact cluster of ascocarps containing as many as forty individuals; ascocarps turbinate, not fully mature in the specimen studied, and in no instance showing dehiscence; the broad, smooth apex rounded and traversed by a shallow furrow, which evidently marks the line of dehiscence; asci (p. sp.) 30–42 × 17–27 μ, 8-spored; ascospores 9–11 μ in diam.

3. CORYNELIA TROPICA (Auersw. & Rab.) Starb. Ark. Bot. **5**: 18–20. 1905.

Endohormidium tropicum Auersw. & Rab. Hedwigia **8**: 89. 1869.

Trullula tropica Sacc. Syll. Fung. **3**: 732. 1884.

Corynelia clavata f. *andina* P. Henn. Hedwigia **36**: 230. 1897.

TYPE: Rab. Fungi Eur. *1261.*

(FIG. 8–12)

Stromata scattered or in linear series along the midrib or margin of the leaf, frequently confluent to form a long, narrow, stromatic cushion; spermogonia and ascocarps borne usually on different stromata; spermogonia globose to irregularly compressed or confluent, densely crowded, forming semiglobose to tubercular masses; young ascocarps conical or irregular from crowding, less uniform in shape in their later stages than in other species, usually becoming characteristically barrel-shaped or short cylindrical, in some individuals indefinitely constricted in the middle zone to give a slightly hour-glass shape, in others retaining the conical form to maturity; when barrel-shaped, the upper end more or less flattened and umbilicate in young stages, the sides marked by a half dozen or more parallel, longitudinal ridges, and the whole surface roughened in such a fashion as to appear rimose; the barrel shape finally lost in many individuals through further development of the upper portion of the ascocarp, which may undergo considerable enlargement and form a broad, somewhat laterally compressed beak terminating in a rather smooth, rounded ridge bearing a transverse groove along its crest; the beak in dehiscence opening widely; the lips sometimes having the brownish, pulverulent appearance typical of the ruptured apex of the ascocarp in other genera; asci 8-spored

except in rare cases of abnormality; ascospores 9–13 μ (commonly 11) in diam.

The original description of the species was based on material collected in South America, at Valdivia, Chile, near the fortieth parallel of south latitude. Though other specimens were later found farther north, the specific name is a misnomer, in that the fungus is less tropical in distribution than any other species of *Corynelia*. It is known, as yet, only from Chile, the Philippine Islands, and New Zealand. Our former diagnosis and the photographs illustrating it, were less satisfactory than those provided for the other species. Most of the specimens which had then been examined were fragmentary and contained few mature ascocarps. Subsequently a number of additional specimens have been studied, including abundant material from New Zealand where the species is common and widely distributed. All the specimens from the Philippines as yet seen by us were collected on *Podocarpus costata*, on Mt. Banahao near Manila. Though a few asci and ascospores are to be found in them most of the ascocarps are immature and marked by the apical umbilicus above mentioned.

4. CORYNELIA OREOPHILA (Speg.) Starb. Ark. Bot. **5**: 18–20. 1905.

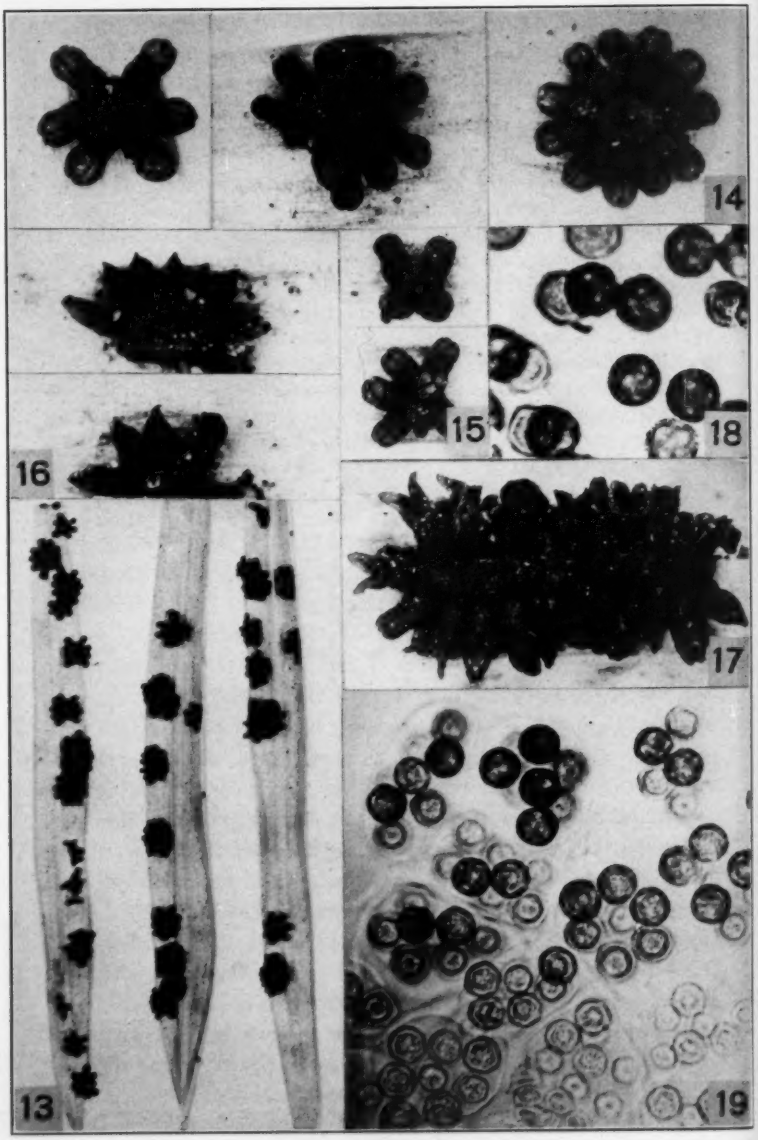
Alboffia oreophila Speg. Anal. Mus. Nac. Buenos Aires **6**: 295 1898.

TYPE: The original material on which Spegazzini based *A oreophila* was collected in 1897 in Argentina on *Podocarpus angustifolia*. The specimen had disappeared before 1920, when Spegazzini sent the writer another collection from Argentina believed by him to be the same fungus. This was found to agree with the material on which Starbäck had based his transfer of the species to *Corynelia*. The latter was collected on the same host in Bolivia, and was deposited as No. *301* in the Herbarium of Robert Fries, at Stockholm (Riksmuseets Botaniska). In addition to these collections, the writer has studied eight others. Three of them were made in Costa Rica. The rest are from South America, the fungus having been seen from Argentina, Chile, Bolivia, Brazil, and Colombia. Various species of *Podocarpus* have been reported as hosts.

(FIG. 25, 26)

Stroma covered with an irregularly arranged group of ascocarps or visible as a prominent, characteristically roughened, sterile cushion, surrounded by a single row of approximately horizontal, radiating individuals as in *C. brasiliensis;* ascocarp characteristic in shape, most closely resembling that of *C. jamaicensis;* the lower half subcylindrical, tapering slightly upward, its surface roughened like the stroma; the upper half smooth to shiny, typically trilobed and trisulcate, in transverse section triangular, tapering slightly downward, giving the ascocarp a somewhat constricted middle zone; the apex subtruncate and centrally depressed; the three furrows united in this depression and running far down the sides between the lobes; dehiscence taking place along the entire length of the furrows making the upper half of the ascocarp deeply tricleft; the three lobes separating and turning back giving a 3-pronged aspect; occurrence of bilobed or quadrilobed individuals rare; ascocarps closed or wedge-shaped in age, as in *C. brasiliensis,* not observed; ascus (p. sp.) 34–42 × 22–30 μ, typically 8-spored; a few 6-spored asci, fewer 5-spored ones, and a single 2-spored one seen, but such variations not correlated with the number of lobes of the ascocarp; ascospores 10–13.5 μ (mostly 12–13) in diam.

The four species, *C. oreophila, C. brasiliensis, C. jamaicensis,* and *C. portoricensis,* are known only from the Western Hemisphere, and are evidently more closely related to each other than to the rest of the genus. In all of them the apex of the ascocarp is definitely lobed, with grooves lying between the lobes and running far down the sides. The two species, *C. oreophila* and *C. brasiliensis,* with typically 8-spored asci, have been found only on the mainland of South and Central America. The other two, *C. jamaicensis* and *C. portoricensis,* with few-spored asci have been reported only from the West Indies. These four clearly had a common ancestry and have probably undergone gradual divergence in morphology as the geographical range has altered. Their development along different lines, from ancestral stock in which the ascocarp was bilobed and the asci 8-spored, is regarded as probable, and this hypothesis was elaborated in our earlier paper. Though admittedly closely related, the four species today are sharply demarcated, and there has been no accumulation of data from later collections to indicate that intergradation between them occurs.



FIGS. 13–19.

5. CORYNELIA BRASILIENSIS Fitzp. Mycologia **12**: 257. 1920.

TYPE: Material collected in the State of São Paulo, Brazil, December, 1896, by Fritz Noack and sent to Elam Bartholomew at Stockton, Kansas, by P. Sydow under the name *C. oreophila*. A portion of the specimen, loaned to us by Bartholomew, was found to be an undescribed species, and was designated as the type of *C. brasiliensis*. Two additional specimens of the same collection in the herbarium of H. Rehm, at Stockholm, were examined. The label on the type specimen states that the collection was made near San Francisco dos Campos, but H. P. Krug of the Instituto Agronomico do Estado de São Paulo at Campinas has written us that no place of that name is known in the State of São Paulo or elsewhere in Brazil. He suggests that S. José dos Campos was perhaps meant. Before the species was described, another collection of material, made by Ule in Brazil and deposited in Rehm's herbarium, was compared with the type and found to be the same. More recently, a third collection was made for us by H. P. Krug (No. *1192*) in São Paulo, at Faz. de Guarda, Campos do Jordão, September 25, 1935, on *Podocarpus Lambertii*. The material, received in extraordinary abundance, shows the fungus in all stages of development. As examination of this collection reveals that the original description was based on aged specimens and gives a false conception of the shape of the ascocarp, the following considerably emended diagnosis, based on the three known collections, is presented.

(FIG. 13–19)

Ascocarps sometimes covering the entire surface of the stroma as a crowded, irregularly arranged group, but more typically confined to its margin, and forming there a single row of horizontal, radiating individuals, bordering a prominent, central, sterile

FIGS. 13–19. *Coryneliа brasiliensis* on *Podocarpus Lambertii*. 13, leaves bearing ascocarps, × 2½. 14, three stromata, each bordered by a row of nearly horizontal, radiating, bilobed ascocarps, × 11. 15, two small clusters of immature ascocarps, each containing a trilobed individual, × 11. 16, stromata bearing aged, closed, wedge-shaped ascocarps, × 11. 17, two crowded stromata, each bordered by a row of ascocarps, × 11. 18, mature ascopores showing echinulate surface and circular lighter-colored areas marking the position of germ-pores, × 730. 19, asci of various ages; spores more mature toward the top, × 510.

cushion; the basal half of the ascocarp considerably roughened and in shape similar to that of *C. oreophila;* the upper half smoother and typically bilobed; trilobed individuals occurring, but less commonly than in *C. portoricensis;* quadrilobed or pentilobed ascocarps not observed; the bilobed individuals similar in gross aspect to those of *C. portoricensis,* but stouter and with less tendency toward lateral compression above; ascocarp in dehiscence opening widely, and, after dissemination of the ascospores, frequently standing wide open and empty, revealing the lighter colored, reddish-brown inner surface of the wall; the recurved lips, however, often tending to come together again so that the emptied ascocarp in age is tightly closed; apex of these aged, closed ascocarps wholly different in shape than before dehiscence, in that the lips fail to regain their former position and instead come together in such a fashion as to form a definite wedge; asci typically 8-spored; ascospores 11–12 μ in diam.

The species is known only from Brazil, and *Podocarpus Lambertii* is the only definitely identified host. Field observations by Krug indicate that attacked plants are smaller and somewhat chlorotic. He collected the fungus at an altitude of about 1500 meters where the host grows most abundantly. He writes: "As you can see in the material sent to you, the perithecia resemble very closely those of *C. portoricensis,* becoming wedge-shaped only in age."

6. CORYNELIA JAMAICENSIS Fitzp. Mycologia **12**: 262. *fig. 6, 7.* 1920.

TYPE: The species was based on material collected, Aug. 10, 1896, by Wm. Harris, on *Podocarpus purdieana,* on Mt. Diablo, Jamaica, near the hotel Holly Mount, and sent to us in 1918 by S. F. Ashby, then Microbiologist of the Department of Agriculture at Kingston. Additional material of the same collection deposited at the New York Botanical Garden as Flora Jamaicensis, No. *6629,* was also seen. Another collection, made in Cuba in 1924, was recently encountered by the writer on material of *Podocarpus* in the phanerogamic herbarium at the New York Botanical Garden.

(FIG. 23, 24)

Stroma bearing several to many ascocarps in a crowded, irregularly arranged group, not exposed to view as a prominent cushion

surrounded by a marginal row of radiating individuals; ascocarp resembling that of *C. oreophila,* though usually somewhat smaller, chiefly trilobed as in that species, but quadrilobed individuals considerably more numerous, and pentilobed ones occasionally found; bilobed ones not yet observed; dehiscence occurring along all the grooves, a quadrilobed individual, for example, having a 4-pronged apex after its rupture; asci (p. sp.) 28–42 × 15–27 μ, mostly 3-spored; the others chiefly 2-spored; asci with more than three spores rare; one containing eight normal spores not yet observed; normal, mature ascospores 11–15 μ in diam.

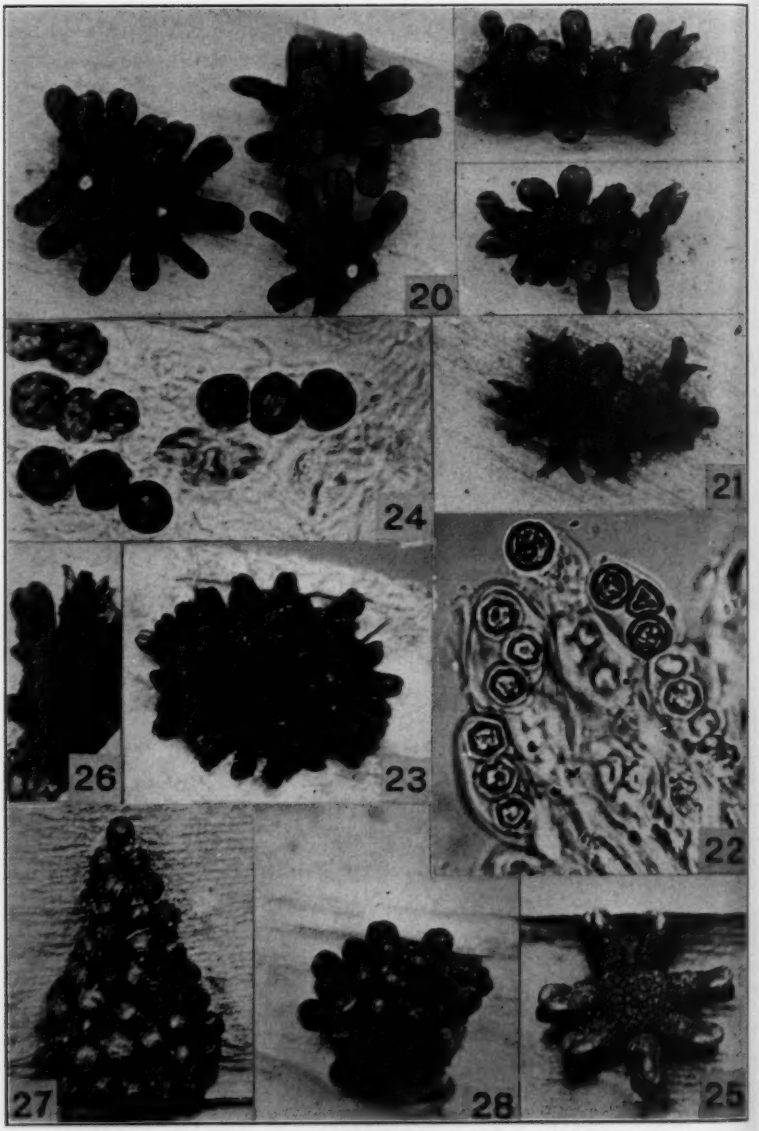
In the possession of typically trilobed ascocarps the species most closely approaches *C. oreophila,* but the tendency toward formation of a larger number of lobes is more pronounced. The characters of the asci and ascospores are essentially the same as in *C. portoricensis,* there being perhaps more 1-spored asci here than in that species.

7. CORYNELIA PORTORICENSIS Fitzp. Mycologia **12**: 259. 1920.
Corynelia clavata var. *portoricensis* Stevens, Ill. Acad. Sci. Trans. **10**: 178–181. 1917.

TYPE: Porto Rican Fungi, No. *784,* received from the herbarium of the University of Illinois, collected, October 20, 1913, near Maricao, Porto Rico, on *Podocarpus coriacea,* by F. L. Stevens. Two other extremely abundant collections of the fungus from Maricao on the same host were studied by the writer before the species was erected. One of these was made, April 2, 1913, by Britton, Stevens, and Hess, the other, March 22, 1916, by Whetzel and Olive. The species is known only from the type locality.

(FIG. 20–22)

Ascocarps chiefly bilobed, but one or more trilobed individuals often centrally placed in the cluster of bilobed ones; quadrilobed ascocarps not yet seen; the trilobed ascocarps very similar to those of *C. oreophila;* the bilobed ones greatly resembling those of *C. brasiliensis,* but somewhat longer and typically more slender, also tending to be more flattened laterally in the upper lobulate portion; the lower part of the ascocarp roughened, subcylindrical, and tapering upward; the upper part smoother, dull to shiny, and tapering downward giving the ascocarp a somewhat narrowed middle zone; the trilobed individuals apically subtruncate and centrally depressed,



FIGS. 20–28.

the three grooves meeting in the depression and running far down the sides between the lobes; the bilobed individuals clavate above, the apex rounded, not angular, crossed by a prominent furrow which runs far down the two broader sides, in some cases extending practically to the base; dehiscence of trilobed individuals as in *C. oreophila,* the upper half of the ascocarp becoming deeply tricleft; dehiscence of bilobed individuals taking place in some cases along the entire length of the groove, the two lobes pulling apart and turning backwards exposing much of the interior of the ascocarp, in other instances occurring only at the apex, a relatively small slit being formed; the lips in neither case observed to close together again in age to form a wedge as in *C. brasiliensis;* asci not differing in the two types of ascocarp, typically 3-spored, frequently 2-spored, and occasionally 1-spored; asci with eight fully formed spores not seen; normal ascospores 10.5–16.5 μ (usually 12–13.5) in diam.; the species in its possession of few-spored asci differing pronouncedly from *C. brasiliensis,* though in gross aspect strikingly similar.

2. TRIPOSPORA Sacc. in Berl. & Vogl. Addit. Syll. Fung. 194. 1886.

Tripocorynelia Kuntze, Revis. Gen. Plant. **3**: 538. 1893.

TYPE SPECIES, *Corynelia tripos* Cooke.

Stromata rounded to elongate, not scattered, arranged in a definite row parallel to the long axis of the leaf, chiefly along the midrib, or similarly along a twig, becoming crowded and confluent, bearing decanter-shaped ascocarps interspersed with irregularly hemispherical spermogonia; ascocarp sessile, composed of a basal flask and a conical to cylindrical beak; apex of beak at maturity perforated and dilated by the extruding ascospores to funnel-form; the mass of spores giving the tip the aspect of an enlarging pul-

FIGS. 20–22. *Corynelia portoricensis.* 20, three clusters of ascocarps; apices chiefly bilobed, but several trilobed, × 11. 1, three clusters of somewhat more mature ascocarps showing dehiscence by transverse cleft along the line of the apical groove; the small, lighter-colored circles at the tips of several individuals being merely high lights in the photograph, × 11. 22, asci and spores, × 510. FIGS. 23, 24. *C. jamaicensis.* 23, a cluster of unruptured ascocarps, chiefly trilobed, a few quadrilobed, × 11. 24, asci with mature spores, × 730. FIGS. 25, 26. *C. oreophila.* 25, pulvinate stroma bordered by a radiating row of almost horizontal ascocarps, × 11. 26, a stroma bearing a dehiscent, three-pronged ascocarp, × 11. FIGS. 27, 28. *C. nipponensis.* Clusters of nearly mature, unruptured ascocarps, some showing transverse apical groove, × 11.

verulent knob, and finally, as the funnel expands, filling it so completely that it becomes a broad convex disc; ascus 8-spored; ascospores characteristic in shape, resembling a caltrop, with 4 (rarely 5) stout, subconic lobes radiating from a rounded central portion, hyaline to light brown when young, becoming darker brown to almost black and opaque at maturity, thick-walled, unicellular, crowded; the radiating lobes closely overlapping to occupy the minimum space in the ascus.

KEY TO SPECIES OF TRIPOSPORA

A. Ascocarp smaller in all its dimensions than in the following species; beak much shorter; ascospores mostly 23–26 μ in diam., with more slender, more uniformly tapering lobes .....1. *T. tripos*
B. Ascospores mostly 26–32 μ in diam.; lobes slightly constricted at the base, broadest at the mid-portion, and tapering abruptly toward the tip .....2. *T. macrospora*

1. TRIPOSPORA TRIPOS (Cooke) Lindau, in E. & P. Nat. Pfl. 1[1]: 413. 1897.

Corynelia tripos Cooke, Grevillea **8**: 34. 1879.

Tripospora Cookei Sacc. in Berl. & Vogl. Addit. Syll. Fung. 194. 1886.

Tripocorynelia tripos Kuntze, Revis. Gen. Plant. **3**: 538. 1893.

TYPE: Specimen in herbarium of Cooke, collected at Cape of Good Hope, S. Africa, near Somerset-East, on *Podocarpus elongata*, by P. MacOwan; compared at Kew by Miss E. M. Wakefield with No. *3150* of Rabenhorst-Winter, Fungi europaei and found to be the same, the latter being *co-type* material. The writer has examined portions of this collection in several institutions and has studied other specimens from Cape Province and Natal.

(FIG. 34–37)

Ascocarps arising usually along the sides of the elongate stromatic cushion; the two rows, with beaks pointing in opposite directions, having a regular and attractive appearance; ascocarp consisting of a globose to ovoidal, roughened, sessile flask, containing the ascigerous locule, and a glabrous, shiny, conical to short-cylindrical beak; apex of beak rounded, blunt, and umbilicate, finally perforate and opening widely to shallow funnel-form; the extruding ascospores filling the funnel to overflowing and forming a convex disc of dark brown, pulverulent aspect, which

equals or somewhat exceeds in diameter the basal portion of the ascocarp; complete dissemination of the spores later exposing the lighter colored inner wall of the funnel, which then appears as a reddish-brown rim bordering the orifice; ascocarp smaller in all its dimensions than in the following species, especially with a much shorter beak; ascospores at maturity dark brown to black and quite opaque, 15–32 μ (mostly 23–26) in diam., measured from the tip of one lobe to that of another, decidedly smaller than in the following species, and differing somewhat in shape, the lobes being more slender and tapering more uniformly from base to apex; species parasitic on the leaves and green twigs of *Podocarpus elongata* and *P. Thunbergii* in South Africa, not known to the writer on other hosts or from other localities; South American collections, earlier included here, now made the basis of the following new species.

2. **Tripospora macrospora** sp. nov.

TYPE: Specimen collected by H. P. Krug, Sept. 25, 1935, at Fazenda da Guarda, Campos do Jordão, Brazil, on *Podocarpus Lambertii* (Herb. Fitzpatrick *2055*). The following additional collections have also been examined; a specimen collected by P. Dusén, at Serrinha, Paraná, Brazil, and sent to us by L. Romell; another from Serra Geral, Brazil, collected by Ule and deposited in the herbarium of Rehm (No. *1744, 1747*); a third collected by S. Venturi, Cerro del Campo, Tucuman, Argentina, and sent to the Missouri Botanical Garden (No. *7725*).

(FIG. 29–33)

Ascocarpus nec forma nec ordine partium a typi distat verum aliquanto longius productus est; rostrum multo longius prostat et flexuosius est; ascospori maiores sunt, diametri 20–36 μ (plurimi vero 26–32), ne forma quidem simillimi; lobi sporidiorum in infima parte paululum constrictiores; in media parte latissimi, in extrema parte subito exiliores fiunt, typo crassiores et rigidiores sunt.

Ascocarp resembling that of the type species, but considerably larger in all its dimensions; beak much longer, more flexuous, and more erect in habit; ascospores larger, 20–36 μ (mostly 26–32) in diam., measured from the tip of one lobe to that of another, and differing from those of the type somewhat in shape; spore lobes slightly constricted at the base, broadest in the mid-portion, tapering abruptly toward the tip, and in general stouter, more rigid, and less graceful than in the type species.

FIGS. 29–37.

3. **Coryneliospora** gen. nov.

TYPE SPECIES, *Capnodium fructicolum* Pat.

Ascocarpus et forma et aperturae specie *Triposporae* par est; ascus octo sporidiorum est; ascospori *Coryneliae* simillimi nec tamen ex omni parte spherici; magnitudine quoque magis variant.

Ascocarp consisting of a sessile flask, tapering upward into a cylindrical beak; apex of beak at maturity perforated and dilated to funnel-form by the extruding ascospores; ascospores echinulate, thick-walled; some large and globose, and essentially identical with those of *Corynelia;* others small, flattened, or irregular.

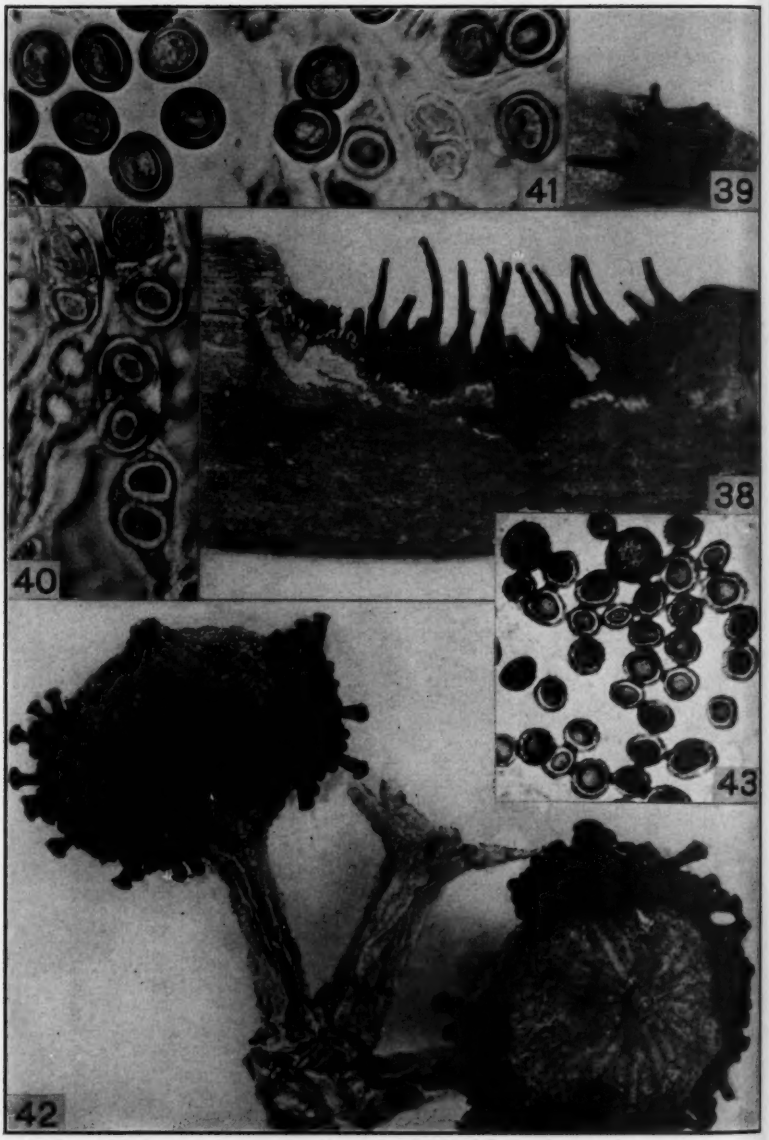
In the form and aspect of the ascocarp the genus closely approaches *Tripospora.* It differs from *Corynelia* chiefly in method of dehiscence, from *Lagenulopsis* in the echinulate character of the spores, and from both of them in that the species does not occur on *Podocarpus.* Our reasons for not recognizing the genus *Lagenula* Arnaud, are elaborated in the second part of this paper in connection with the discussion of *Caliciopsis.*

1. **Coryneliospora fructicola** (Pat.) comb. nov.

Capnodium fructicolum Pat., Jour. de Bot. **3**: 258. 1889.
Corynelia carpophila Syd., Engler Bot. Jahrb. **45**: 264. 1910.
Corynelia fructicola (Pat.) v. Höhnel, Sitzber. Kais. Akad. Wiss. **120**: 450. 1911.
Lagenula fructicola (Pat.) Arn. Ann. Epiphy. **16**: 269. 1930.

TYPE: Original material of *Capnodium fructicolum,* in herbarium of Patouillard at Harvard University, on fruits of *Myrsine sp.*

FIGS. 29–33. *Tripospora macrospora.* 29, elongate stromata, bearing ascocarps on leaves of *Podocarpus,* nat. size. 30, mature ascocarps, × 11. 31, ruptured ascocarps, the funnel-shaped apex of each filled with a pulverulent mass of extruding ascospores, × 11. 32, mature, free ascospores, × 180. 33, two asci, showing the compact arrangement of the eight stellate ascospores, × 730. FIGS. 34–37. *T. tripos.* 34, a row of erumpent stromata bearing a double row of ascocarps, × 11. 35, mature ascocarps as seen in lateral view, the funnel-shaped apex of each packed with extruding ascospores and having the aspect of a convex disc, × 11. 36, mature ascocarps, the apex of each appearing as a shallow funnel; the reddish-brown, inner surface surrounding a narrow throat still filled with brownish-black ascospores, × 11. 37, nearly mature ascospores, of smaller size and somewhat different shape from those of the preceding species, × 180.



FIGS. 38–41. *Lagenulopsis bispora.* 38, stromata on leaf of *Podocarpus* bearing spermogonia and ascocarps, × 11. 39, cluster of ascocarps showing

collected in southwestern China, in the Province of Yun-nam, by Delavay. The type specimen of *Corynelia carpophila* Sydow, collected by Lane Poole in the Transvaal, South Africa, on *Rapanea melanophleos* is the same fungus. Other specimens from South Africa on this host and one from India on *Myrsine africana* have been seen.

(FIG. 42, 43)

Stromata fructicolous, rounded, reaching 1 mm. in diam., commonly confluent to form a crust which sometimes completely encircles the fruit; surface of young stroma covered with densely crowded spermogonia, among which the ascocarps develop and protrude as radiating spines; spermogonium globose, provided with a prominent, apical, perforate papilla, and tapering at the base into a more or less evident stalk; spermatia hyaline, rod-shaped to allantoid, 4–6 × 1 μ; ascocarp decanter-shaped; the basal flask tapering upward into a cylindrical beak; apex of beak blunt, marked in early stages by a tiny umbilicus, later perforated and dilated by the extruding spores to shallow funnel-form; spore-mass forming an enlarging, pulverulent knob, which finally broadens into a thick, convex disc; complete dissemination of the spores exposing the lighter colored, reddish-brown, inner surface of the funnel; asci (p. sp.) 20–25 × 11–14 μ, 8-spored; ascospores echinulate, thick-walled, brown, varying considerably in size and shape; large spherical spores, 10.5 μ in diam.

4. **Lagenulopsis** gen. nov.

TYPE SPECIES, *Corynelia bispora* Fitzp.

Ascocarpus lecythi forma, sessilis, in exile rostrum exiens; apertura non differt a *Caliciopsidis;* ascus duorum sporidiorum; ascospori globosi vel subglobosi, muris crassissimis, levibus, fulvis.

Stromata rounded, typically crowded, bearing radiating, spine-like ascocarps interspersed with smaller, inconspicuous spermogonia; mature ascocarp composed of a subconical, sessile flask, containing the ascigerous locule, and a long, slender beak; apex of beak in dehiscence perforated and dilated by the extruding ascospores to funnel-form; the funnel narrow and inconspicuous as in *Caliciopsis,* not broad and shallow as in *Tripospora* and *Corynelio-*

the brown, apical knobs formed in dehiscence, × 11. 40, two-spored asci; the ascospores of various ages, × 730. 41, mature, free, extremely thick-walled ascospores, × 730. FIGS. 42, 43. *Coryneliospora fructicola.* 42, fruits of *Rapanea melanophleos* bearing spermogonia and ascocarps on crowded to confluent stromata, × 11. 43, mature ascospores, with the plane of focus raised to show the echinulate surface, × 730.

spora; the spore-mass forming an inconspicuous, pulverulent brown knob; ascospores globose to subglobose, with an exceptionally thick, smooth, brown wall.

Differing from *Caliciopsis* in the much larger and far thicker-walled spores, in the typically sessile ascocarp, and in the occurrence on *Podocarpus;* allied by this host relationship with *Tripospora* and *Corynelia.*

1. **Lagenulopsis bispora** comb. nov.

Corynelia clavata f. *macrospora* Sydow, Adolf Friedrichs Deutsche Zentral-Afrika-Exped. 1907–1908. **2**: 100. 1910.
Corynelia bispora Fitzp. Mycologia **12**: 242. 1920.

TYPE: Original material of *C. clavata* f. *macrospora* collected on *Podocarpus milanjianus* Rendle, deposited as No. *2547* in herbarium of Sydow, at Berlin.

(FIG. 38–41)

Stromata erumpent on the upper or lower surface of the leaf or emergent along its edge, in the latter case apparently bordering wounds made by chewing insects; spermogonium globose to subconical, apically umbilicate, finally perforate; spermatia hyaline, yellowish in mass, fusiform, 5–8 × 2 μ; ascocarp 1–2 mm. in length; ascus apparently constantly 2-spored; ascospores 11–15 μ in diam.

Sydow erected *Corynelia clavata* f. *macrospora* on a single collection of material received from central Africa in 1908. The writer examined a fragmentary portion of this collection, found the fungus to be actually very different from *C. clavata,* and erected on it the new species *C. bispora.* Attention was called to the 2-spored nature of the ascus, and to the resemblance of the ascocarp in shape and method of dehiscence to that of *C. fructicola.* As we had never seen another specimen of the fungus, it was especially pleasing to receive an ample amount of material in 1933 collected in Jamaica in July of the preceding year by Professor Duncan S. Johnson. The above emended diagnosis is based chiefly on this Jamaican material. The asci and ascospores are indistinguishable from those of the type, and the fructifications agree in size and shape.

DEPARTMENT OF PLANT PATHOLOGY,
CORNELL UNIVERSITY,
ITHACA, NEW YORK